Elucidating the virulence control network in Francisella tularensis

Elucidating the virulence control network in *Francisella tularensis*

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Abstract

The Gram-negative bacterium *Francisella tularensis* is the causative agent of tularemia and a model intracellular pathogen. It is also considered a potential bioweapon, as *F. tularensis* is highly infectious and has the potential to cause fatal disease in humans. Many factors required for *F. tularensis* virulence have been identified, yet we know relatively little regarding how these factors are regulated at the level of transcription. In order to further understand the regulation of virulence factors in *F. tularensis*, we have systematically determined the genomic regions associated with all of the transcription factors implicated in virulence using chromatin immunoprecipitation coupled with high-throughput DNA sequencing (ChIP-Seq).

We have determined that three transcription factors critical for intracellular growth and survival, MglA, SspA, and PigR, are found at virtually all promoters and that PigR, a putative DNA-binding protein, requires MglA in order to specifically associate with promoters. We have identified a DNA sequence found in promoters positively regulated by PigR, which may constitute a PigR-responsive element. Our results indicate that PigR appears to function solely as a positive regulator in combination with the complex formed by MglA and SspA, but that MglA (as part of the MglA-SspA complex) appears also to function as a negative regulator of gene expression, working independently of PigR to negatively regulate σ^{32}-controlled genes.

By correlating the locations of virulence-associated transcription factor ChIP-Seq peaks with nearby genes, we have predicted potential regulons under the control of each factor and, in

the case of OxyR and the LysR-type transcription regulator FTL_0040, we have determined which putative regulon members have been implicated as virulence factors. We have determined that the virulence-associated transcription factor PmrA may promote expression from genes on the *Francisella* pathogenicity island indirectly. We have found that three proteins, PmrA, IclR, and the LysR-type transcription regulator FTL_1568, associate with regions of the genome that are not typical for canonical transcription regulators; we propose that they may function in part to organize the chromosome as nucleoid-associated proteins. Finally, we have generated a model of the network of transcription factors that control virulence in *F. tularensis.*

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The journey through graduate school is not short and there have been so many influences along this path; they have shaped me into the person I am now, and I am grateful for them all.

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To my parents,

Kenneth and Karen Levasseur,

for all their love and support

Chapter 1

Introduction

Introduction

The disease tularemia was first characterized in 1911, after an outbreak of a plague-like disease was observed in small rodents in Tulare County, CA, USA (McCoy and Chapin, 1912). The causative bacterium was isolated and originally named *Bacterium tularense* (McCoy, 1911). Later, the genus of the causative bacterium was renamed *Francisella* in honor of Dr. Edward Francis, a physician and member of the United States Public Health Service (Dorofe'ev, 1947; Jellison, 1972). Dr. Francis both named the disease and performed much of the pioneering work on tularemia, although it should be noted that in some literature from the mid-20th century, *F. tularensis* was occasionally referenced as *Pasteurella tularensis* (Jellison, 1972).

F. tularensis subspecies and strains

There are four subspecies of *F. tularensis*, all of which are capable of causing disease, albeit with significant variability in severity (Ellis et al., 2002): *tularensis*, *holarctica*, *mediasiatica*, and *novicida.*

Strains of *F. tularensis* subsp. *tularensis*, which are also referred to as type A strains, are the most pathogenic of the subspecies (Evans, 1985). Fewer than ten *F. tularensis* subsp. *tularensis* bacteria can constitute an infectious dose, and in humans there are reports of mortality rates for untreated disease up to 60% (Tärnvik and Chu, 2007). *F. tularensis* subsp. *tularensis* is endemic to North America (Keim et al., 2007). The strain most frequently used by laboratories to study *F. tularensis* subsp. *tularensis* is the sequenced strain SCHU S4 (Larsson et al., 2005).

F. tularensis subsp. *holarctica*, also known as type B strains, are the other cause of tularemia in humans, although the disease caused by these strains is significantly milder and not thought to be life-threatening (Keim et al., 2007). *F. tularensis* subsp. *holarctica* is found in both North America and Eurasia (Keim et al., 2007). The former Soviet Union used *F. tularensis* subsp. *holarctica* to attempt to generate a vaccine strain, starting in the 1930s (Tärnvik and

Berglund, 2003). They were successful in generating several strains attenuated for virulence in humans, and one was transferred to the United States in 1956 (Ellis et al., 2002). This strain, called the Live Vaccine Strain (LVS, hereafter referred to as *F. tularensis* LVS) is frequently used as a model to study *F. tularensis* pathogenesis as cells of this strain are still lethal to mice (Eigelsbach and Downs, 1961). All the studies performed on *F. tularensis* in this work have been performed with *F. tularensis* LVS.

The *F. tularensis* subsp. *mediasiatica* is reported to both have a similar lethal dose in animal models and cause disease in humans similar to *F. tularensis* subsp. *holarctica* (Larsson et al., 2009; Olsufjev and Meshcheryakova, 1983), but is only found in an isolated geographical region in Central Asia (Oyston et al., 2004). As a result, there are few studies performed on this particular subspecies.

Originally classified as its own *Francisella* species, *F. tularensis* subsp. *novicida* is now generally considered a subspecies of *F. tularensis* based on its genetic similarities with the other subspecies (Santic et al., 2006), although we will continue to refer to it in this work as *F. novicida*. *F. novicida* does not cause disease in immunocompetent humans, although the disease it causes in animal models is very similar to the disease caused by the human pathogens (Ellis et al., 2002; Hollis et al., 1989). Thus, the *F. novicida* sequenced strain U112 is frequently used as a model for *F. tularensis* subsp. *tularensis*.

Tularemia

Infection by *F. tularensis* can occur via a number of routes. Contact with, or ingestion of, contaminated food, water, or infected animal tissue can all cause disease (Sjöstedt, 2007). Arthropod vectors, which include fleas, ticks, biting flies, and mosquitoes, can transmit *F. tularensis* (Petersen et al., 2009). Additionally, *F. tularensis* is easily aerosolized and infection can occur from inhalation of particulate matter from infected carcasses or even dust from contaminated soil, grain, or hay (Sjöstedt, 2007).

The disease caused by exposure to *F. tularensis* generally has an incubation period of 3-5 days, although it may extend up to 21 days (Tärnvik and Berglund, 2003). The disease generally manifests with flu-like symptoms; fever, chills, fatigue, general body aches, headaches, and nausea are all commonly reported (Evans et al., 1985; Tärnvik and Chu, 2007). The type of disease and additional symptoms vary dependent upon the site of infection. The two most common types of tularemia are ulceroglandular and respiratory (Nigrovic and Wingerter, 2008).

Exposure through a break in the skin, commonly via an infected biting arthropod, causes ulceroglandular tularemia. In this case, a papule develops at the site of infection that generally becomes a pustule over the course of several days (Evans et al., 1985). As this occurs, lymph nodes become swollen and enlarged. While the initial ulcer generally heals even without treatment, the lymph nodes can continue to enlarge and potentially become suppurative (Tärnvik and Berglund, 2003). Ulceroglandular tularemia is similar in the case of infection with either type A (*F. tularensis* subsp. *tularensis*) or type B (*F. tularensis* subsp. *holarctica*) strains.

Respiratory tularemia occurs after inhalation of aerosolized bacteria, and symptoms and outcome are significantly different upon infection by type A or type B strains (Tärnvik and Berglund, 2003). Type B strains generally result in mild, non-fatal respiratory disease. Respiratory tularemia with a type A strain, however, is an acute, severe disease with variable flu-like symptoms including those listed above, high fever, cough, and dyspnoea (difficulty breathing) (Evans et al., 1985; Tärnvik and Berglund, 2003). Without antibiotic treatment, mortality rates of 30-60% are reported (Dennis et al., 2001). With treatment, however, mortality rates are less than 2% (Dennis et al., 2001).

Treatment of *F. tularensis* is typically successful with antibiotics, specifically aminoglycosides, tetracyclines, and more recently, quinolones (Tärnvik and Chu, 2007). Development of antibiotic-resistant *Francisella* within patients is not a significant concern, as it is

not a member of the commensal flora and there is no evidence that it can be spread via human-to-human contact.

Classification as a potential bioweapon

Not only is *F. tularensis* extremely infectious, it also has a history of development as a bioweapon, in the 1930s by the Japanese biological warfare program (Harris, 1992), and later by both the United States and the former Soviet Union (Dennis et al., 2001). The United States cancelled their biological weapons program and signed the Convention on the Prohibition of the Development, Production and Stockpiling of Bacteriological (Biological) and Toxin Weapons and their Destruction in 1972 (Oyston et al., 2004). Given its history as a developed bioweapon, its highly infectious nature, the large number of transmissible routes, and its potential to cause mortality, the United States government considers *F. tularensis* a potential bioweapon.

Intracellular lifestyle

As an intracellular pathogen, *F. tularensis* must be able to enter the cell, survive the hostile eukaryotic intracellular environment, replicate, and escape to continue the infection. While *F. tularensis* is competent to infect a number of different cell types, the macrophage appears to be a critical niche of infection for subsequent dissemination (Councilman and Strong, 1921; White et al., 1964). A critical function of macrophages is to phagocytose and destroy potential pathogens. However, *F. tularensis* has co-opted this function in order to gain access to the cell. *F. tularensis* promotes its own phagocytosis by inducing formation of a looping pseudopod which eventually engulfs the bacterium (Clemens et al., 2005) (Figure 1.1). Most phagocytosed vacuoles would merge with early, and then late, endosomes, become acidified, and fuse with lysosomes. The environment inside the vacuole is toxic and the inactivated contents would be recycled. Experimental evidence thus far suggests that the fate of the *F. tularensis*-containing phagosome (FCP) depends on the state of the bacteria prior to uptake. If

the *F. tularensis* were opsonized prior to phagocytosis, the FCP does not become acidified and the bacteria escape into the cytoplasm between 2-4 hours post-infection (Clemens et al., 2009; Jones et al., 2012). If the *F. tularensis* were unopsonized when phagocytosed, the FCP becomes transiently acidified and the bacteria escape into the cytoplasm in less than 1 hour (Jones et al., 2012; Santic et al., 2007). Once in the cytoplasm, the bacteria replicate to high numbers and, after 24-72 hours, trigger apoptosis of the cell, escaping to continue infection (Belhocine and Monack, 2012; Lai et al., 2001).

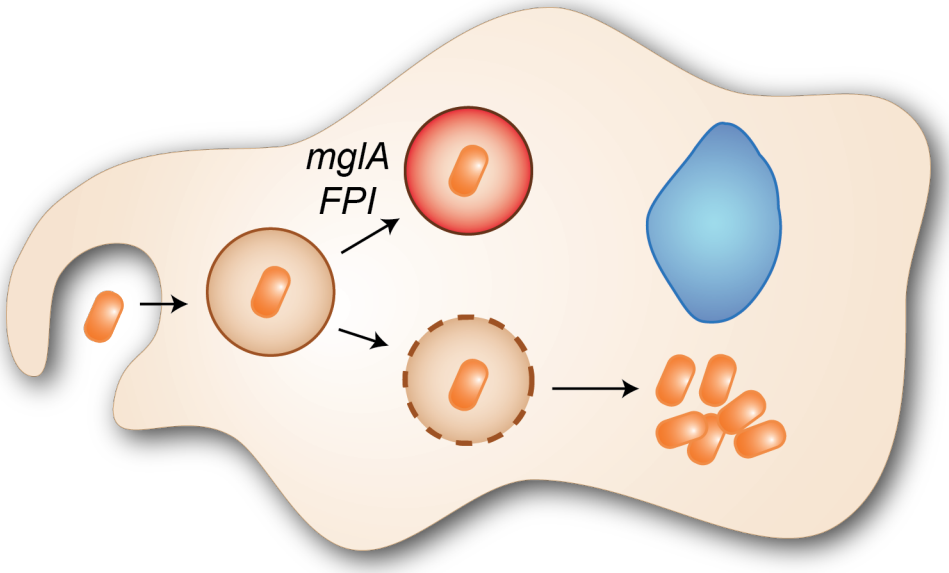

Figure 1.1. *F. tularensis* is an intracellular pathogen. Diagram depicts the intramacrophage growth of *F. tularensis*. Bacteria are engulfed via looping pseudopods and enter a *Francisella* containing particle (FCP, circle with solid brown line). Bacteria are able to escape the FCP (circle with dashed brown line) and replicate in the cytosol, eventually triggering cell death to escape the macrophage. However, bacteria with mutations in *mglA* or bacteria missing the FPI cannot escape the FCP and are degraded (circle with sold black line). Diagram adapted from Pechous et al., 2009.

The *Francisella* Pathogenicity Island

The *Francisella* Pathogenicity Island (FPI) is a genetic region encompassing approximately 25-30 kb of the *F. tularensis* genome and encodes two putative operons (Barker and Klose, 2007). A genetic screen performed in *F. novicida* first identified two genes located on the FPI as critical for intramacrophage growth and survival (Gray et al., 2002). That the screen was performed in *F. novicida* was likely critical to the early identification of genes present on the FPI; while the genomes of *F. tularensis* subsp. *tularensis* and *holarctica* contain two identical copies of the FPI, *F. novicida* only contains one, which allowed the FPI to be easily identified by inactivating mutagenesis (Nano et al., 2004).

The region that comprises the FPI is likely to have been horizontally acquired, as the GC-content of the region is extremely low in comparison to the low GC-content of the rest of the genome (27% vs 32%) (Nano and Schmerk, 2007). There are 16 genes which comprise the conserved core of the FPI (Figure 1.2), all of which are found in both *F. tularensis* LVS and sequenced *F. tularensis* subsp. *holartica* clinical strains (Bröms et al., 2010). With the exception of *pdpE*, all of the FPI genes have been demonstrated to be important for virulence in animal models (Bröms et al., 2010). Upstream of *iglA, F. tularensis* subsp. *tularensis* and *novicida* contain two additional genes, *pdpD* and *anmK* (Bröms et al., 2010). It has been shown that the *pdpD* gene in *F. novicida* is critical for virulence, and that the presence of the *anmK* gene, which is furthest upstream, is also critical, although possibly only because its promoter may be used for driving expression from the rest of the operon (Ludu et al., 2008).

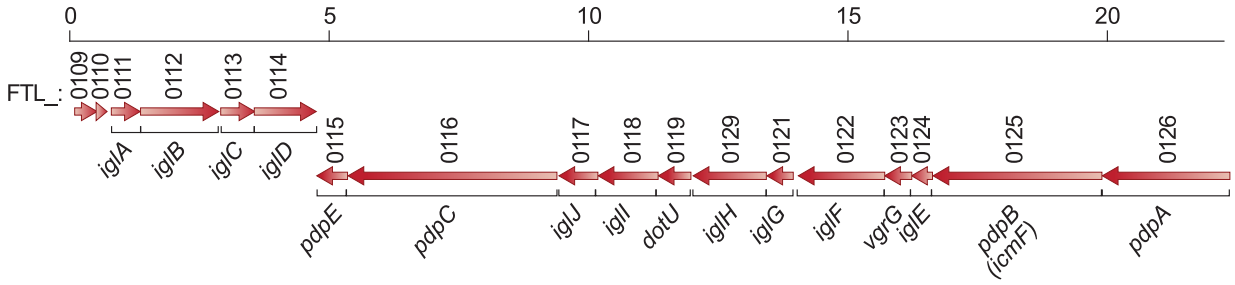

Figure 1.2. The *Francisella* Pathogenicity Island in *F. tularensis* LVS. The scale is indicated at the top in kb.

The FPI locus is required for intramacrophage survival and growth, and it appears to be critical specifically for arresting phagosome maturation and escape of the bacteria into the cytosol. As many recent studies have indicated that the FPI genes likely encode a type VI secretion system (T6SS) (Barker and Klose, 2007; Barker et al., 2009; Brotcke et al., 2006; Bröms et al., 2011; de Bruin et al., 2011; 2007; Lindgren et al., 2013; Nano et al., 2004; Santic et al., 2007), this most likely occurs via translocation of bacterial effector proteins to directly modulate host cell pathways. However, the homology between the FPI and other described T6SSs is limited; the FPI appears to be only distantly related to other well-studied T6SSs (Barker et al., 2009; Bingle et al., 2008; de Bruin et al., 2007). Recently, the crystal structure of a putative structural T6SS protein from *F. novicida*, DotU, was reported and despite the lack of strong similarity at the amino acid level, it does appear to have significant structural similarities to DotU homologs in other organisms (Robb et al., 2012). Experiments in *F. tularensis* LVS have recently indicated that eight proteins encoded on the FPI, specifically IglE, IglC, IglI, VgrG, PdpE, PdpA, IglJ, and IglF, are secreted into macrophages, dependent on the presence of the FPI-encoded IglG and DotU proteins (Bröms et al., 2012). It is not known if these secreted proteins constitute effectors and many questions remain regarding the structure and function of the FPI-encoded proteins in promoting intracellular survival.

RNA polymerase in *Escherichia coli* and *F. tularensis*

Not only is transcription an essential process, but it is also a critical point of regulation for most bacterial virulence programs. Transcription occurs through the activity of the highly conserved DNA-dependent RNA polymerase (RNAP) enzyme, which is capable of synthesizing RNA. In *E. coli*, the core RNAP consists of multiple subunits: two α subunits, a β subunit, a β' subunit, and an ω subunit. The core enzyme contains all of the catalytic machinery required to initiate RNA synthesis, but in order to initiate transcription at specific promoters, core RNAP must associate with the σ subunit, which together are called the RNAP holoenzyme.

F. tularensis is unique in that it encodes genes for two distinct α subunits (Charity et al., 2007); all other bacterial species examined to date contain only one *rpoA* gene (which encodes the α subunit) and incorporate a homodimer of α subunits into RNAP. In *F. tularensis*, both α subunits are incorporated into RNAP *in vivo* (Charity et al., 2007). *In vitro* data is consistent with both subunits incorporated as heterodimers (Mukhamedyarov et al., 2011). While the genes that encode the two α subunits, *rpoA1* and *rpoA2*, are reported to be potentially essential genes (Gallagher et al., 2007), several transposon mutant screens have identified *rpoA2* as a factor critical for virulence, suggesting that *rpoA2* may not be essential (Asare et al., 2010; Su et al., 2007).

For most σ factors, binding to the RNAP core enzyme (forming the holoenzyme) induces a conformational change in σ such that a specific DNA-binding domain is unmasked. RNAP holoenzyme is then competent to recognize, bind, and initiate transcription at promoters recognized by RNAP containing the σ factor. Most bacteria encode not only a so-called housekeeping σ factor, but also several others, termed alternative σ factors, which are used to respond to specific stimuli and initiate the appropriate transcriptional response. It has been proposed that the number of σ factors present in a bacterial species increases in relation to both its genome size and the complexity of its environment (Pérez-Rueda et al., 2009; van Nimwegen, 2003). *F. tularensis* has a broad host range yet its genome is quite small, encoding about 2,000 genes in approximately 1.9 Mb. It contains two genes for σ factors: *rpoD*, which encodes the housekeeping factor σ^{70}, and *rpoH*, encoding σ^{32}, or the heat-shock σ factor (Grall et al., 2009).

The structure of bacterial promoters

Promoter regions contain specific sequences of DNA that allow for σ factor recognition and subsequent initiation of transcription. These DNA sequences contribute to what is called the basal promoter strength, or the frequency with which transcription occurs at the promoter region

in the absence of other factors. For many promoters, the majority of the basal activity is determined by two 6 bp regions, called the -10 and -35 elements, which are so named due to their location, approximately 10 and 35 bp upstream from the transcription start site. The σ subunit makes direct contact with these regions of DNA in a sequence-specific manner, such that -10 and -35 elements that are closer to the consensus sequence will have more σ binding and greater basal promoter strength. While most σ subunits are part of the diverse σ^{70} family and utilize the -10 and -35 elements for promoter recognition, the σ^{54} family members are structurally different; these alternative σ factors recognize specific sequences at -12 and -24 regions, which are located 12 and 24 bp upstream of the transcription start site, respectively (reviewed in Österberg et al., 2011).

Basal promoter activity is also affected by a number of other factors, including the spacing between the -10 and -35 elements and the presence of additional points for RNAP holoenzyme contact, which could include a discriminator region, an extended -10 element, or an UP element. The discriminator region is found between the transcription start site (also referred to as +1) and the -10 element (Travers, 1980). In *E. coli* and *Thermus aquaticus*, the presence of additional guanines in the discriminator region improves binding of the major σ factor to promoters (Feklistov et al., 2006; Haugen et al., 2006). The extended -10 element consists of an additional recognized dinucleotide sequence 1 bp upstream of the -10 element (Barne et al., 1997), found frequently in *E. coli* promoters with suboptimal -10 or -35 regions (Hook-Barnard et al., 2006; Mitchell et al., 2003) and commonly used in *Bacillus subtilis* (Camacho and Salas, 1999). The UP element is an approximately 20 bp region typically found in *E. coli* a few bp upstream from the -35 element (Gourse et al., 2000; Ross et al., 1993). In *E. coli*, the consensus UP element is very AT-rich and its presence significantly increases transcription, providing another point of contact between DNA and part of RNAP, the C-terminal domain of the RNAP α subunit (αCTD) (Gourse et al., 2000).

The consensus sequences for the described promoter elements have been determined in *E. coli*, but their similarity to their counterparts in *F. tularensis* is unknown. There is significant conservation between the *E. coli* σ^{70} and the *F. tularensis* σ^{70} in the regions critical for DNA contact, suggesting that the consensus sequences are likely to be similar in these two organisms. It should be noted, however, that the canonical elements that *E. coli* promoters consist of are AT-rich. The entire genome of *F. tularensis* is very AT-rich, which makes identifying specific promoter regions, in the absence of other data, more challenging.

σ factor competition

Each bacterial species encodes at least one housekeeping σ factor, which is thought to be both the most abundant and have the highest affinity for core RNAP during normal growth. Alternative σ factors can recognize and bind to DNA sequences that are different from the housekeeping σ factor recognition sites, so they are often used to specifically regulate particular promoters. Alternative σ factors can be used to respond to specific stimuli, via altering the transcriptional program to preferentially express genes required to adapt to the given stimulus. An example would be the *E. coli* σ^{32} factor, the heat-shock σ factor. The mRNA encoding σ^{32} is present at low temperatures, but the translation start site is occluded by complementary base pairing, preventing protein synthesis. Under heat stress, the base pairing that prevents the synthesis of σ^{32} is destabilized, thus increasing the abundance of σ^{32} (Morita et al., 1999). Subsequently, part of the σ^{32}-mediated response to heat stress is involved in preventing the increase in temperature from damaging DNA; σ^{32} upregulates genes involved in maintaining genome integrity, such as the recombination repair proteins *recA* and *recJ* (Nonaka et al., 2006).

In *E. coli*, the amount of core RNAP enzyme in each cell is thought to remain relatively constant, providing a limited pool of core RNAP for association with the available σ factors (Grigorova et al., 2006; Piper et al., 2009). The association between core RNAP and a σ factor

is dependent on both the affinity of a particular σ factor for core RNAP as well as the abundance of the available σ factor. Essentially, σ factors must compete for association with a limited amount of core RNAP. Thus, much of the regulation with respect to which σ factor is used can be attributed to increasing or decreasing the concentration of a σ factor, which is in fact a highly regulated process (Österberg et al., 2011). In the absence of specific signals for alternative σ factor use, there are a number of dedicated factors, termed anti-σ factors, which are responsible for interacting with and sequestering specific σ factors (reviewed in Helmann, 1999). But regulating the abundance of an available σ factor can be used in concert with another strategy to allow alternative σ factor use, such as specifically promoting the formation of RNAP containing a particular σ factor by utilizing a protein that interacts with a σ factor such as Crl.

The protein Crl was identified based on its ability to promote transcription from σ^{38}-dependent promoters in *E. coli* (Pratt and Silhavy, 1998). It has subsequently been found to achieve this effect by interacting directly with σ^{38} and apparently promotes the formation of σ^{38}-containing RNAP holoenzyme (Gaal et al., 2006; Banta et al., 2013). This appears to effectively allow even low concentrations of the alternative sigma factor σ^{38} to associate with core RNAP when in competition with other σ factors (Gaal et al., 2006; Banta et al., 2013). Crl sets the precedent in the literature for a transcription factor that appears to specifically and directly promote formation of a particular RNAP holoenzyme species to control gene expression.

Regulators of virulence gene expression in *F. tularensis*

MglA and SspA

MglA and SspA are two proteins that have been found to be critical for *F. tularensis* intracellular survival and replication (Baron and Nano, 1998; Charity et al., 2007). Both are members of the stringent starvation protein A (SspA) family, classified based on homology to the *E. coli* transcription factor SspA. In *E. coli*, SspA is a protein produced during the stringent response, the response to amino acid starvation (Williams et al., 1994). The stringent response

is characterized by decreased transcription from rRNA promoters, limited protein production, and increased abundance of the small molecule ppGpp (guanosine 3',5'-bisdiphosphate, with its precursor pppGpp [collectively referred to here as (p)ppGpp]) (reviewed in Jin et al., 2012). Despite the significant decrease in protein production, the amount of SspA present in the cell increases during the stringent response (Williams et al., 1994). The crystal structure of the SspA homolog in *Yersina pestis* has been determined and SspA protein was found as a homodimer (Hansen et al., 2005). It has been shown that SspA, in a complex with a co-activator protein Lpa, can associate with σ^{70}-containing RNAP to positively regulate the expression of bacteriophage P1 late genes, although the mechanism by which this occurs is unclear (Hansen et al., 2003). No putative DNA-binding domains are present in SspA and none were reported in the crystal structure for the *Y. pestis* homolog of SspA (Hansen et al., 2005). Thus, it seems likely that the effect of SspA on P1 late gene expression is due to its ability to associate with RNAP holoenzyme and possibly interact with Lpa, rather than by binding DNA itself.

MglA and SspA in *F. tularensis* can interact to form a heteromer and MglA and SspA have been shown to directly interact with σ^{70}-containing RNAP, likely as a heteromer (Charity et al., 2007). The regulons of MglA and SspA are extensive and virtually identical, and include approximately 100 genes (Brotcke et al., 2006; Charity et al., 2007; Guina et al., 2007). MglA/SspA regulated genes include many virulence-related genes as well as many which are, thus far, unlinked to any virulence phenotype (Brotcke et al., 2006; Charity et al., 2007; Lauriano et al., 2004). Among the genes whose expression is most significantly promoted by MglA and SspA are genes on the FPI (Brotcke et al., 2006; Charity et al., 2007). MglA and SspA are critical for FPI gene expression, and the decrease in FPI gene expression undoubtedly contributes to the failure of *mglA* or *sspA* mutants to survive intracellularly and contributes to the avirulence of *mglA* and *sspA* mutants in mice (Baron and Nano, 1998; Brotcke et al., 2006; Charity et al., 2007; Lauriano et al., 2004; Nano et al., 2004). The mechanism by which MglA and SspA influence gene expression, however, remains unclear.

PigR and (p)ppGpp

A protein that interacts with the MglA-SspA complex and also regulates a common set of genes is called PigR in *F. tularensis* LVS (FevR in *F. novicida)* (Brotcke and Monack, 2008; Charity et al., 2009). PigR contains a putative helix-turn-helix (HTH) motif and shows homology to members of the MerR family of transcription regulators (Brotcke and Monack, 2008; Charity et al., 2009).

PigR contains a putative HTH motif; this putative HTH motif is similar in sequence and relative location to the HTH motif found in MerR family proteins (Brotcke and Monack, 2008; Charity et al., 2009). The conservation of a similar putative DNA-binding domain between PigR and MerR family members suggests that PigR may bind DNA and may be a member of the MerR family. MerR family members contain two domains: one domain that contains a highly conserved winged HTH motif and another more variable C-terminal domain, which is generally the effector-binding domain (Brown et al.). The variable C-terminal domains frequently function via binding of co-activators, and a subfamily of MerR regulators (including MerR itself) uses this region to specifically interact with and respond to metals (Summers, 2009). The mechanism for promoter activation by MerR family members appears to be largely conserved. Target promoters identified for canonical MerR family members generally have longer, suboptimal spacing (19 or 20 bp) between the -10 and -35 promoter regions. MerR family members frequently associate with the promoter region in both the inactive and active form. Upon activation, a shift in protein conformation occurs which results in a dramatic bending and unwinding of promoter DNA, effectively shortening the distance between the -10 and -35 elements to allow for efficient holoenzyme recognition (Brown et al.; Newberry and Brennan, 2004).

The HTH motif in PigR was identified by comparison with the conserved domain cd01104 in the NCBI CDD database. The founding member of this group is the *E. coli* MerR

family member MlrA. MlrA and the other proteins with domains found in the cd01104 group can be distinguished from other MerR family members in their lack of an obvious dimerization domain, which would typically be found as part of the N-terminal domain after the HTH motif. Additionally, studies of MlrA have indicated that it may have a mechanism of promoter activation that differs from other MerR family members. MlrA has been found to bind upstream from the -35 region (from -146 to -113) of its well-described target promoter, *csgD* (Ogasawara et al., 2010). Despite the lack of an obvious dimerization domain within the protein, the identified DNA-binding site consists of a palindrome, suggesting that MlrA could function as a dimer (Ogasawara et al., 2010). Although the homology between PigR and MerR family members is significant, there is as of yet no evidence that PigR functions as a MerR family member by directly binding DNA to influence transcription.

It has been found, however, that PigR interacts with RNAP via association with the MglA-SspA complex (Charity et al., 2009). This interaction appears to be influenced by (p)ppGpp (Charity et al., 2009). The production of (p)ppGpp in both *E. coli* and *F. tularensis* is dependent on two enzymes with (p)ppGpp synthetase activity, RelA and SpoT (Charity et al., 2009). Cells that lack PigR or that lack both RelA and SpoT exhibit reduced FPI gene expression and are unable to cause disease in mice (Charity et al., 2009).

Given that PigR, MglA, and SspA all regulate a common set of genes, we have previously hypothesized that PigR is a site-specific DNA-binding protein critical for directing MglA-SspA associated RNAP to target promoters and activating transcription by making contacts both with the DNA and with the RNAP-associated MglA-SspA complex (Charity et al., 2009). In turn, (p)ppGpp is thought to exert its effects, either directly or indirectly, by promoting the interaction between PigR and the RNAP-associated MglA-SspA complex (Figure 1.3).

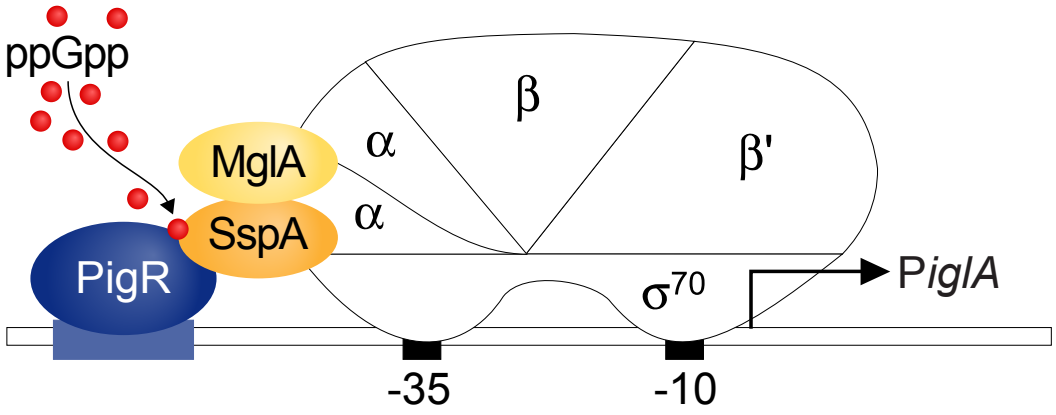

Figure 1.3. A model for the regulation of target gene expression by PigR, the MglA-SspA complex, and (p)ppGpp. PigR, a putative site-specific DNA-binding protein, interacts directly with the RNAP-associated MglA-SspA complex at target promoters. The presence of (p)ppGpp is thought to stimulate this interaction, either directly or indirectly. Figure adapted from Charity et al., 2009.

PmrA

PmrA is the only protein encoded by *F. tularensis* LVS with significant similarity to a two-component system response regulator, although the genomes of *F. tularensis* SCHU S4 and *F. novicida* encode an additional putative response regulator (FTT1543 and FTN_1453, respectively) (van Hoek, 2013). Two-component systems are typically encoded by two genes within the same operon and consist of a sensor kinase and a response regulator. Canonically, the sensor kinase responds to an environmental stimulus and alters the phosphorylation state of the response regulator. The response regulator then binds to promoter sequences of regulated genes to induce or repress transcription (Stock et al., 2000). Sensor kinases and response regulators that are not found together in operons are frequently referred to as orphans. These orphan sensor kinases and orphan response regulators may have specific partners, they may be able to interact with several partners, or they may have atypical interactions that influence gene expression in other ways (reviewed in Raghavan and Groisman, 2010).

In *F. tularensis* LVS, there are three putative sensor kinases annotated, none of which are found in the same putative operon as *pmrA*; thus, all the annotated sensor kinases or response regulators are considered orphans. However, it should be noted that two of the annotated sensor kinases, FTL_1878 and FTL_1879, appear to be incorrectly annotated. The sequences of FTL_1878 and FTL_1879 together encode one protein, homologous to the sensor kinase annotated as *kdpD* in *F. novicida* and *F. tularensis* subsp. *tularensis*. In the *F. tularensis* subsp. *holarctica* species, *kdpD* contains a premature stop codon and the resulting fragments are annotated as two separate sensor kinases (Alkhuder et al., 2010). Thus, it is likely that the two annotated sensor kinases FTL_1878 and FTL_1879 that together correspond to *kdpD* are not functional sensor kinases. This leaves one potentially functional sensor kinase in *F. tularensis* LVS, encoded by *qseC* (FTL_1762).

Previous studies have demonstrated a link between the transcription factor PmrA and virulence (Bell et al., 2010; Mohapatra et al., 2007; Sammons-Jackson et al., 2008). Not only

was a *pmrA* mutant attenuated for replication in macrophages and for virulence in a mouse infection model, it was found that PmrA positively regulates a subset of genes controlled by MglA, SspA, and PigR, including *pigR* itself (Mohapatra et al., 2007; Sammons-Jackson et al., 2008). Additional work in *F. novicida* indicated that purified PmrA could bind its own promoter, as well as the promoter of the FPI gene *pdpD*, which is present in the *F. tularensis* subsp. *tularensis* SCHU S4 and *F. novicida* U112 strains but not in any *F. tularensis* subsp. *holarctica* strains, including LVS (Bell et al., 2010).

It is also reported that KdpD is likely to be the cognate sensor kinase for PmrA (Bell et al., 2010). However, as previously mentioned, LVS does not appear to contain an intact *kdpD* gene. The effect of PmrA on virulence gene expression does not appear to be strain specific, as cells of LVS that contain a deletion of *pmrA* exhibit a similar downregulation of *pigR* and other MglA/SspA/PigR regulated genes when compared to wild-type cells. Moreover, a LVS *pmrA* mutant is also attenuated for virulence in an animal model (Sammons-Jackson et al., 2008). Thus, if PmrA functions in a canonical manner to alter gene expression after phosphorylation by a sensor kinase, it appears that QseC, the only potentially functional sensor kinase in LVS, would be the likely candidate to interact with PmrA.

Several other groups have studied PmrA in *F. novicida*, under the alternative name of QseB, in the context of biofilm formation (Durham-Colleran et al., 2010; Zogaj and Klose, 2010). A *pmrA* deletion strain in LVS was found to be attenuated for biofilm formation (Durham-Colleran et al., 2010). Additional work in *F. novicida* found PmrA may affect biofilm formation through altering the expression of *F. novicida*-specific genes responsible for controlling bis-(3'-5')-cyclic dimeric GMP (cdi-GMP) (Zogaj and Klose, 2010). However, no homologs of these genes, encoding proteins with either diguanylate cyclase or phosphodiesterase activity, have been identified in either *F. tularensis* subsp. *holarctica* or *F. tularensis* subsp. *tularensis* strains. In fact, as biofilm formation phenotypes correlated inversely with virulence phenotypes, it was suggested that loss of the ability to generate cdi-GMP is a factor associated with the increased

virulence of *F. tularensis* subsp. *holarctica* and subsp. *tularensis* strains in comparison to *F. novicida* (Zogaj et al., 2008).

IclR

The IclR family of transcription factors is defined by an N-terminal HTH domain and a region of conservation in the C-terminus that may bind to a regulatory substrate (Krell et al., 2006). Additionally, IclR family members are also defined by the conservation of their overall size, ranging from 240-280 amino acids. These family members are widely found and have been identified in over 60 genera of bacteria and archaea (Krell et al., 2006). By both promoting and/or repressing expression of their target genes, IclR family members also appear to regulate a wide variety of cellular processes, including those for metabolic pathways, resistance to small molecules, determinants of pathogenicity, and developmental processes such as sporulation (Molina-Henares et al., 2006).

An *F. tularensis* IclR protein was identified as being important for virulence in a screen that assessed the ability of *F. novicida* transposon mutants to infect the spleen after intraperitoneal injection (Weiss et al., 2007). The mutant containing a transposon insertion in the *iclR* gene was severely attenuated for infection, but in a cell culture model appeared to stimulate macrophage cell death faster and induced higher levels of a specific proinflamatory cytokine than wild-type *F. novicida* (Weiss et al., 2007). When used to infect macrophages lacking components of the cell death pathway, the mutant containing a transposon insertion in the *iclR* gene failed to both promote cell death and induce levels of the specific proinflamatory cytokine (Weiss et al., 2007). This suggests that the IclR protein in *F. tularensis* subsp. *novicida* is important for the regulation of factors that modulate the host immune response in order to promote infection.

The effect of IclR on the virulence of *F. tularensis* appears to be strain-specific, however. A study performed on the *F. tularensis* LVS homolog of IclR found it to be dispensable for

replication in macrophages and virulence in animals (Mortensen et al., 2010). Similarly, deletion of the *F. tularensis* subsp. *tularensis* gene encoding the IclR homolog had no effect on virulence in a mouse model, but the authors were able to replicate the attenuation phenotype of the *F. novicida iclR* mutant (Mortensen et al., 2010). Many of the genes found to be regulated by IclR in *F. tularensis* LVS appear to be pseudogenes. The *F. tularensis* subsp. *tularensis* homologs of the IclR-regulated genes are also pseudogenes, but the *F. novicida* homologs appear to encode functional proteins (Mortensen et al., 2010). This suggests that there are subspecies-specific virulence pathways, and IclR is dispensable for virulence in the subspecies that cause human disease.

Fur

The ferric uptake regulator, Fur, is a site-specific DNA-binding protein that typically functions as a repressor. Fur binds Fe^{2+} directly, and in its Fe^{2+}-bound state represses transcription of target genes under conditions considered iron-replete (Bagg and Neilands, 1987). Fur binds to a specific sequence on the DNA, termed a Fur box (Baichoo and Helmann, 2002; de Lorenzo et al., 1987), and prevents transcription by occluding RNAP from the promoter region of the target gene. When the cellular concentration of iron is reduced, Fur dissociates from regulated promoters to allow transcription (Bagg and Neilands, 1987).

In *F. tularensis*, Fur has been shown to regulate the *fsl* operon, which encodes a siderophore used in ferric iron uptake, in response to iron availability (Buchan et al., 2009; 2008; Ramakrishnan et al., 2012). Two Fur boxes are found associated with the operon; one Fur box is found upstream of *fslA*, and the other Fur box is located upstream of *flsE*, which encodes the receptor for the siderophore (Deng et al., 2006; Kiss et al., 2008; Ramakrishnan et al., 2008).

Multiple studies have linked iron concentration to virulence gene expression in *F. tularensis* (Buchan et al., 2008; 2009; Deng et al., 2006; Lenco et al., 2005). Many other bacteria increase virulence gene expression upon sensing low iron conditions, consistent with

the idea that the intracellular environment in host cells is iron-limited (Andrews et al., 2003; Caza and Kronstad, 2013). In *F. tularensis*, it has been demonstrated that the levels of FPI-encoded proteins IglC and PdpB, as well as a number of FPI-encoded transcripts, are increased when cells are grown in low iron conditions (Deng et al., 2006; Lenco et al., 2005). The increase in protein and transcript abundance that appears to occur under conditions of low iron is suggestive of regulation by Fur; thus, the promoter regions of affected genes on the FPI were inspected for putative Fur boxes, which were identified (Deng et al., 2006).

To test the effect of the Fur protein on the expression of FPI genes, one of the FPI genes with increased transcription under low iron conditions, *iglB*, was examined using a reporter construct. While expression of an *iglB-lacZ* reporter increased in response to low iron, it was not repressed by overexpression of Fur, unlike an *fslC-lacZ* reporter (Buchan et al., 2008; 2009). Another gene in the Fur-regulated *fsl* operon, *fslA*, was examined in the context of both low iron conditions and in cells with an insertion mutation in the *fur* gene. In both low iron conditions and in cells with a mutation in *fur*, *fslA-lacZ* expression increased, consistent with the repression of *fslA* by Fur in iron-replete conditions (Buchan et al., 2009). Expression of another FPI gene reporter construct, *iglA-lacZ*, showed moderate increases in low iron conditions. However, expression of the *iglA-lacZ* reporter also continued to increase in response to low iron conditions in a *fur* mutant background (Buchan et al., 2009). This suggests that the promoter driving expression of *iglA* and *iglB* may respond to iron abundance independently of Fur. There are no studies published to date regarding other genes potentially regulated by Fur in *F. tularensis*.

OxyR

OxyR is a LysR-type transcription regulator used to mediate the cellular response to oxidative stress—specifically the presence of hydrogen peroxide (reviewed in Imlay, 2013). The effect of OxyR as an activator or a repressor depends upon the spacing between the OxyR

binding site and the promoter elements. OxyR binds DNA as a homotetramer, and when reduced, binds across five helical turns, with a bend in the DNA in the third turn. Under conditions of oxidative stress, OxyR is directly oxidized and undergoes a conformational change such that the homotetramer binds across four consecutive helical turns, occupying a slightly different DNA binding site (Choi et al., 2001; Toledano et al., 1994).

In *E. coli*, OxyR regulates the expression of close to 40 genes (Chiang and Schellhorn, 2012; Zheng et al., 2001). The OxyR regulon includes the *oxyR* gene itself, which OxyR negatively regulates (Christman et al., 1989; Warne et al., 1990), and the *oxyS* gene which is transcribed divergently from the *oxyR* gene, encodes a regulatory RNA, and is positively regulated by OxyR (Altuvia et al., 1997). The presence of OxyR is conserved among diverse bacterial species, and OyxR positively regulates the expression of genes encoding catalases (such as the hydroperoxidase I, encoded by *katG*) and alkyl hydroperoxidase reductase (such as Ahp, encoded by *ahpCF*) in many gammaproteobacteria. Catalases and Ahp are used to detoxify H_2O_2 into H_2O and O_2; their regulation by OxyR provides a direct link between sensing and detoxifying H_2O_2 (reviewed in Imlay, 2008).

In *F. novicida*, OxyR was identified as critical for virulence in a screen of transposon insertion mutants that used *Drosophila melanogaster* as an infection model (Moule et al., 2010). The screen also identified genes encoding DNA repair enzymes, suggesting that responding to DNA-damaging oxidative stress is a critical aspect of successful infection. In the *D. melanogaster* model, a specific immune response is responsible for generating reactive oxygen species; in flies lacking this immune response, the *F. novicida* mutant containing a transposon insertion in the *oxyR* gene was able to successfully survive within the fly (Moule et al., 2010).

Chromosomal organization and nucleoid-associated proteins (NAPs)

While the issues surrounding chromosome organization and compaction have been the subjects of investigation in eukaryotes for some time, interest in the manner in which bacterial

chromosomes are organized has recently increased. Early electron micrograph images prompted the theory that bacterial DNA compaction might mostly be achieved though DNA supercoiling, in contrast to the histone-mediated organization found in eukaryotes (Kellenberger, 1988, reviewed in Dame, 2005). While it is clear that supercoiling is critical for DNA organization and compaction, it has become widely recognized that proteins not only play a role in generating and stabilizing supercoiled DNA, but also in the organization of DNA. A major difference between eukaryotic and prokaryotic DNA organization is that prokaryotic DNA is generally accessible to the transcription machinery. Some have suggested that the entire genome in prokaryotes is available and competent for transcription (Wade et al., 2005), although there are studies suggesting that this is not universally true (Dorman, 2004; Myers et al., 2013). Regardless, the prokaryotic chromosome appears to be organized in a way that allows accessibility yet still maintains an appropriate level of compaction.

Evidence for chromosomal organization and plasticity specifically related to transcription comes from studying the stringent response in *E. coli*. Ribosomal RNA operons are among the most highly transcribed during normal cell growth in *E. coli*, and fluorescence microscopy of epitope-tagged RNAP subunits reveals distinct foci (reviewed in Jin et al., 2012). These discrete foci, termed transcription foci or transcription factories, correspond to rRNA operons and the many molecules of RNAP involved in their transcription. During the stringent response, however, growth slows, transcription from rRNA and tRNA operons dramatically decreases, and the transcription foci are no longer present, suggesting a significant shift in chromosomal organization (Jin et al., 2012).

A functional class of proteins, nucleoid-associated proteins (NAPs), has been described which, critically, are able to alter the trajectory of DNA via bending, wrapping, or bridging DNA molecules (Dillon and Dorman, 2010). Given the nature of their interaction with DNA, it is not surprising that many NAPs affect transcription. And since NAPs appear to bind DNA at many, sometimes extended, sites throughout the chromosome, it is also unsurprising that many are

abundant proteins. Well-described NAPs that fit this description, i.e., affect the trajectory of DNA, influence transcription, and are abundant, include the proteins H-NS, HU, Fis, and IHF (reviewed in Dame, 2005; Dillon and Dorman, 2010). However, the functional class of NAPs has extended to include a number of proteins that may or may not be highly abundant and may be implicated in a number of other cellular processes. These include MukB, Lrp, CbpA, the *Borrelia burgdorferi* protein EbfC (whose *E. coli* homolog is YbaB), Dps, Crp, and the *Mycobacterium tuberculosis* protein EspR (Blasco et al., 2012; Dillon and Dorman, 2010; Grainger et al., 2005; Jutras et al., 2012). It should be noted that in *F. tularensis*, the only recognizable NAP identified thus far is the homolog of HU, encoded by *hupB*.

Including a protein that has been well-studied as a canonical transcription factor, such as Crp, in the NAP category is indicative that the line between transcription factor and NAP is becoming increasingly blurred and the distinction less meaningful. The effect of NAPs on transcription varies; some, like members of the H-NS family are involved in silencing gene expression (Dorman, 2004), while others, like EspR, are critical in promoting virulence gene expression (Raghavan et al., 2008). Gene regulation by NAPs has been shown to occur, even for one protein, via a number of mechanisms. Fis has been shown to repress transcription both by binding at target promoters and occluding RNAP, as well as by preventing the isomerization of promoter open complexes (Schneider et al., 1999). IHF can function to promote the contacts between the αCTD of σ^{54}-containing RNAP and target promoters (Macchi et al., 2003) and IHF can physically bend DNA to allow contact between other regulators and RNAP (Santero et al., 1992). Additionally, the activity of specific transcription factors may be influenced by the presence of NAPs; the *E. coli* protein FNR is found to be limited to binding sites which are not occupied by H-NS, IHF, or Fis (Myers et al., 2013).

Recent research into the structure of the bacterial chromosomes suggests that the overall organization of the chromosome may be dynamic, driven largely by supercoiling and the location of highly-expressed genes (Fisher et al., 2013; Le et al., 2013). Bacterial NAPs appear

to play a more significant role at a more local level of organization, in regions of less than 100 kb in *Caulobacter crescentus* (Le et al., 2013). The protein abundance of many NAPs changes with growth state, so the nature of their association with DNA must change accordingly. Additionally, use of proteins such as Crp and Lrp in chromosomal organization would allow the dynamics of organization to change in response to nutritional cues.

Chromatin immunoprecipitation followed by high-thoughput sequencing (ChIP-Seq) in bacteria

It is possible to identify all of the regions at which a given protein associates with the genome by performing chromatin immunoprecipitation followed by high-throughput sequencing, or ChIP-Seq. The first step of this process is to crosslink the protein-protein and protein-DNA complexes present in the cell; the crosslinking can be achieved by adding a crosslinking agent such as formaldehyde. The protein of interest is then specifically immunoprecipitated from cell lysates, isolating the protein-DNA interactions that were present at the time of crosslinking. The next steps are to purify the fragments of DNA that were isolated as a result of their association with the protein of interest, generate libraries of immunoprecipitated DNA for high-throughput sequencing, and sequence the DNA. Subsequent computational steps are aimed at identifying which regions of the genome are more frequently immunoprecipitated with the protein of interest than some background control. These steps include aligning the short sequenced DNA reads to the genome and determining criteria for significant enrichment of reads. Typically, a set fold-enrichment of ChIP-Seq reads over background is used. Another criteria that is indicative of specific association between a protein and a region of DNA is sometimes referred to as positive peak shift (Figure 1.4).

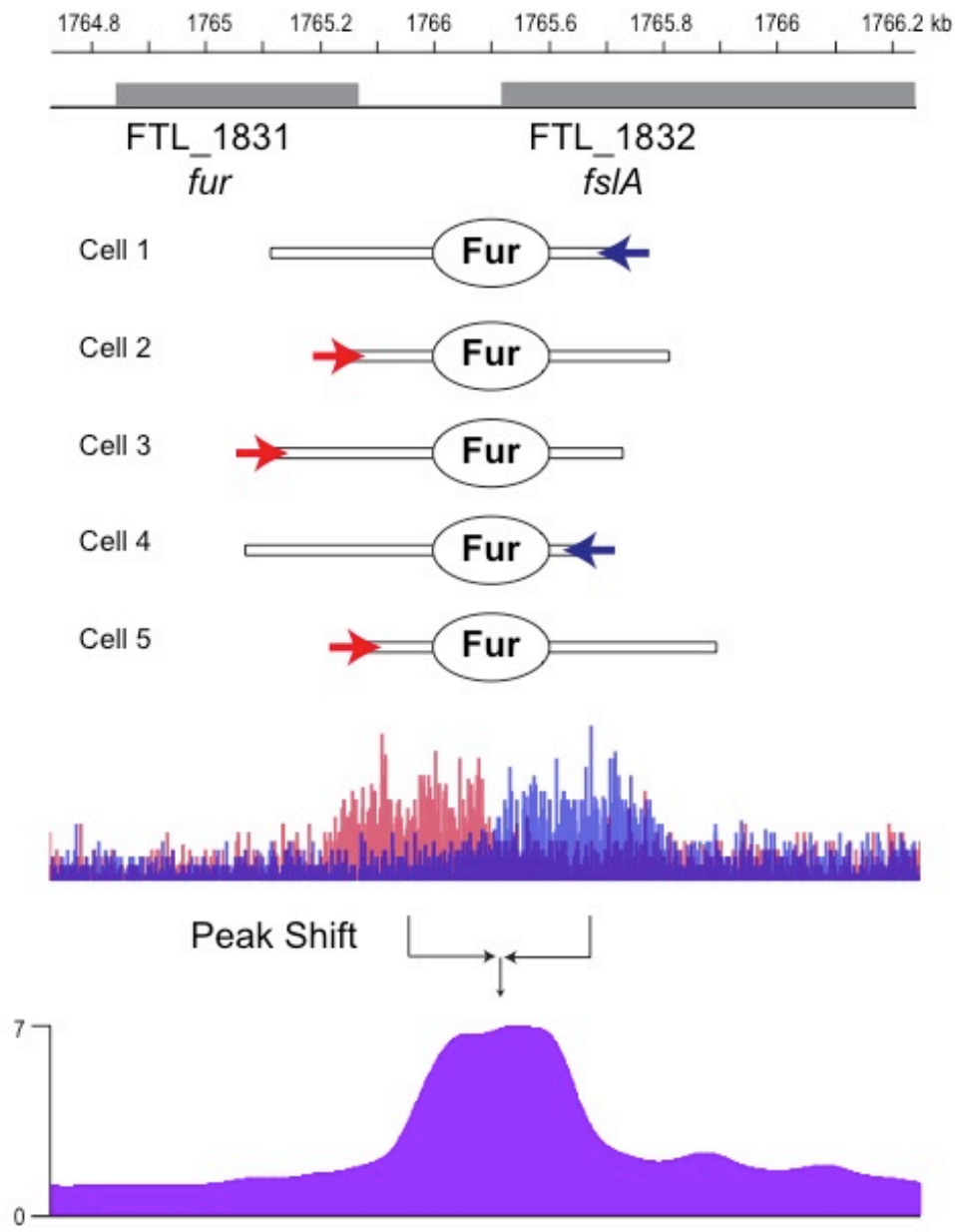

Figure 1.4. Peak shift in ChIP-Seq data is indicative of protein association. The schematic diagram depicts the visualization of ChIP-Seq data from Fur (from this study; see Chapter 3). Genomic location is listed along the X-axis at the top, in both kb and locus number. Fragments of DNA associated with Fur protein from different cells are variable in length. Each DNA fragment will be sequenced from the 5′ end, resulting in local enrichment of strand-specific reads on either side of the Fur-binding site (red indicates sequencing from the plus strand, blue indicates sequencing from the minus strand). The overlapping red and blue bars indicate actual read data. The point of maximum read enrichment along each strand is identified and the maximum points for the two strands are merged midway, yielding a normalized peak representative of Fur association, depicted in purple. Figure adapted from Valouev et al., 2008.

A peak shift is essentially an artifact that results from the association of a protein with a region of DNA. The protein of interest will have been associated with fragments of DNA of various sizes and the length of the sheared DNA on either side of the associated protein will be variable (Figure 1.3). In sequencing a large number of fragments, the location of the protein should average out to approximately the center of a given region with more reads than background. However, sequence data is always generated directionally, from the 5′ end to the 3′ end of DNA fragments. For bona fide regions of protein association, this will result in a large number of reads aligning to the positive strand upstream from the site of association and a large number of reads aligning to the negative strand downstream from the site of association (Figure 1.3). Thus, the association of a protein with a single region of the DNA results in two strand-specific peaks of enrichment. To more accurately represent the location of the DNA-associated protein, the two strand-specific peaks can be merged at the mid-way point between the two peaks. A positive peak shift refers to shifting these two peaks together in a manner that reflects association of a protein with the region of DNA (i.e., reads on the positive strand shifting downstream and reads on the negative strand shifting upstream; Figure 1.3). Negative peak shift, which would reflect an abundance of reads aligning to the negative strand followed by reads aligning to the positive strand, could not be generated by the immunoprecipitation of DNA between the two local peaks and so is indicative of artifactual enrichment.

The use of ChIP-Seq has several advantages in comparison to the earlier-developed method of chromatin immunoprecipitation followed by the hybridization of immunoprecipitated DNA to a microarray, called either ChIP-chip or ChIP-on-chip (Park, 2009). When using ChIP-Seq, direct sequencing of the immunoprecipitated DNA circumvents the need to design a high-density microarray containing the entire genome sequence. And because the output signal no longer relies on hybridization of DNA to probes of pre-determined sizes, it is possible to obtain more accurate and more refined peak sizes. Additionally, the dynamic range of sequencing is much larger than the dynamic range of microarray technology, resulting in greater sensitivity.

The use of ChIP-Seq in bacterial studies as a method to identify locations of protein association within the genome has become common; while from 2009-2012, fewer than ten papers studying bacteria included ChIP-Seq analyses, at least thirteen papers used ChIP-Seq to study bacterial transcription factors in 2013. Published studies range from identification of the binding sites for a single transcription factor, to large-scale studies such as the work performed by the Galagan group, which has already assessed 50 transcription factors in *M. tuberculosis* by ChIP-Seq and aims to examine all of the *M. tuberculosis* transcription factors using this approach (Galagan et al., 2013a; 2013b).

Several initial ChIP-Seq studies in bacteria have been performed on well-described NAPs (Galagan et al., 2013a; Kahramanoglou et al., 2011; Prieto et al., 2011; Wang et al., 2013). The protein binding regions for proteins such as IHF, H-NS and Fis have been previously assessed by ChIP-chip (Castang et al., 2008; Cho et al., 2008; Grainger et al., 2006). However, comparison between datasets has demonstrated that ChIP-Seq results in significantly better resolution in terms of binding site identification (Kahramanoglou et al., 2011).

In studying proteins that have been considered canonical transcription factors (i.e., those that function by binding to specific sites present at the promoters of target genes to influence gene expression), an apparently frequent result is the identification of protein-association sites at non-canonical, that is, non-promoter, regions (Bush et al., 2013; Butcher et al., 2011; Jungwirth et al., 2013; Perkins et al., 2013; Scott et al., 2013; van Kessel et al., 2013). Occasionally, these regions are discarded from analyses (Bush et al., 2013; Perkins et al., 2013; Scott et al., 2013). In some cases, the investigators choose to validate the unexpected association between the protein and DNA, as in the case of the transcription factor GlxR in *Corynebacterium glutamicum*; GlxR was found to bind a number of intragenic sequences, and one that was unlikely to be near a promoter was shown to bind purified GlxR by electrophoretic mobility shift assay (EMSA) (Jungwirth et al., 2013).

These apparently non-canonical binding sites need not necessarily be truly non-canonical. Intragenic binding sites could be representative of unidentified promoters, be representative of alternative promoters, be representative of promoters for small RNA species, or reflect poor genome annotation and actually are within intergenic regions.

Alternately, non-canonical binding sites could represent transcription factors functioning at sites distal to the regulated promoter. There are a number of examples of long-range interactions in prokaryotes, such as repression of the *E. coli araBAD* operon. The AraC protein binds a site over 200 base pairs from the *araBAD* promoter in order to repress transcription (reviewed in Schleif, 2003). However, while these long-range interactions are not artifacts and do affect the biology of the cell, it typically seems that increasing the distance between the regulator's site of binding and the regulated promoter tends to result in a diminished regulatory effect. If the occupancy of non-canonical sites do have regulatory consequences, it is possible that the regulated promoter is distant and thus the effect of the regulator is slight, which could contribute to the reason these binding sites were not previously identified.

Another explanation for proteins that associate with both canonical and non-canonical sites may be that they function analogously to the recently characterized EspR protein in *M. tuberculosis*. EspR was originally identified as a transcription factor necessary for promoting expression of genes at the *espA* locus, encoding a secretion system critical for virulence (Raghavan et al., 2008). More recently, EspR was found to both make long-range DNA looping interactions to directly promote *espA* expression, as well as play a role in organizing chromosome structure (Blasco et al., 2012; Hunt et al., 2012). This suggests that transcription factors may have multiple functions when associated with DNA, and the category of NAP may continue to expand. These newly classified NAP members may or may not be highly abundant and may function both as site-specific transcription factors and chromosomal organization modulators, rather than be highly abundant and associate with the entire chromosome.

Many ChIP-Seq studies in bacteria have also identified a large number of sites detected with low enrichment over background, or so called weak binding sites (Chumsakul et al., 2011; Galagan et al., 2013a; 2013b; Jungwirth et al., 2013). However, the extent to which a region is enriched by ChIP-Seq can be influenced by a large number of factors and does not necessarily correlate to binding strength. Additionally, choices regarding the significance or cut-off values for signal intensity are frequently based on arbitrarily chosen fold enrichments. Several studies suggest that widespread, apparent weak binding may not be entirely artifactual.

Analysis of FNR binding sites across the *E. coli* genome identified significant variability in enrichment of ChIP-Seq peaks (Myers et al., 2013). The authors found that, under the conditions examined, the FNR binding sites with variable enrichment were saturated *in vivo*. By examining the relative enrichment of FNR binding sites with a range of induced FNR protein, it was determined that the variation in ChIP-Seq peak enrichment was likely due either to variable crosslinking efficiency or variable immunoprecipitation within given genomic contexts (Myers et al., 2013). These results highlight that comparing the relative enrichment of a particular factor at different locations may not be biologically relevant.

Other studies of widespread identification of ChIP-Seq peaks with low enrichment suggest that protein abundance may be a key determinant in identifying regions of association for a particular factor. It logically follows that under conditions of low protein abundance, it will be harder to detect low-affinity binding sites. The detection of more putative protein binding sites in the presence of more protein has been reported in work performed in *M. tuberculosis* (Galagan et al., 2013a), suggesting that the sites of association for these factors are not saturated. It was found that increasing the abundance of the transcription factor of interest results in the identification of more peaks and, generally, with greater enrichment of the previously identified peaks (Galagan et al., 2013a). Additionally, genes associated with many of the weaker peaks have been found to be regulated by the transcription factor of interest,

suggesting that the peaks identified with weak enrichment may be biologically relevant (Galagan et al., 2013a).

One way to circumvent the issue of protein abundance is to induce saturation of binding sites. Saturation of binding sites was the goal of Davies et al; in their work they overexpressed the *Vibrio cholerae* Fur protein in order to identify all possible binding sites (Davies et al., 2011). Using this strategy, known sites of Fur association were significantly increased and no false positives were reported. Many of these sites were linked to Fur regulation by RNA-Seq studies, and inspection of intragenic binding regions identified novel putative small RNAs under the control of Fur (Davies et al., 2011).

Another study provides an alternate suggestion for the function of sites with weak binding. In a paper demonstrating a novel approach to ChIP-Seq, Genome Footprinting by high-throughput sequencing (GeF-Seq), essentially DNaseI footprinting *in vivo* followed by high-throughput sequencing, the authors identify a specific bipartite binding motif for the transcription factor AbrB (Chumsakul et al., 2011). They had found a large number of weak binding sites, but based their analysis and motif identification on a subset which corresponded to the most enriched sites. Subsequently, they re-assessed the weaker sites for the AbrB bipartite motif and found that a single motif, rather than the bipartite motif, was enriched at the weaker sites. They suggest that the sites with weak affinity may be used to concentrate AbrB along the DNA to allow it to localize to high affinity sites (Chumsakul et al., 2011).

A significant strength of ChIP-Seq studies is their ability to identify regulatory networks, which would be difficult to discern using traditional transcriptomic approaches. By using ChIP-Seq to identify the direct targets of a particular transcription factor, and coupling that information with transcriptomic analysis such as RNA-Seq, regulatory networks and hierarchies can be revealed. An example is that ChIP-Seq of a single transcription factor, such as ArcA in *E. coli*, reveals that ArcA is in the center of a significant regulatory network, regulating other transcription factors and thus both directly and indirectly affecting the expression of a wide

range of genes (Park et al., 2013). The information gleaned from the fifty transcription factors examined by ChIP-Seq in *M. tuberculosis* thus far is starting to elucidate the complex network of transcription regulation used in bacterial life (Galagan et al., 2013a).

Summary

F. tularensis is an important human pathogen which requires appropriate gene regulation for pathogenesis. While the transcription factors MglA, SspA, PigR, and PmrA, along with the small molecule (p)ppGpp are necessary for intramacrophage survival and virulence, there are many other transcription factors which contribute to regulating *F. tularensis* virulence. ChIP-Seq is a powerful tool that can be used to identify direct targets of transcription factors, as well as elucidate protein function and reveal regulatory networks.

In the following chapters, I report my studies aimed at finding the regions of the *F. tularensis* genome that are bound by those transcription factors implicated in controlling virulence. In doing so, I determine that MglA, SspA, and PigR are found at essentially all promoters, suggesting that PigR is not a site-specific DNA-binding protein that is only found at target promoters. I also identify differences in the regulatory activities of the MglA-SspA complex and PigR. I find that PmrA is likely to indirectly affect regulation of genes found on the FPI through its direct effect on the expression of the *pigR*. I additionally identify putative NAPs in *F. tularensis*, characterize regulons of all the virulence-associated transcription factors, and uncover a regulatory network among transcription factors.

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Chapter 2

The key virulence regulators MglA, SspA, and PigR are found essentially at all promoters in *Francisella tularensis*

Author contributions: All of the work in this chapter was performed by Kathryn Levasseur.

Abstract

In *Francisella tularensis,* MglA, SspA, and PigR are transcription factors that are essential for the ability of the bacterium to replicate within macrophages. Previous work has established that MglA and SspA form a heteromeric complex that interacts with RNA polymerase (RNAP). An additional interaction between the MglA-SspA complex and PigR, a putative DNA-binding protein, is thought to be necessary to specifically promote expression of virulence genes. We determined those regions of the *F. tularensis* genome that associate with MglA, SspA, and PigR using ChIP-Seq and found that these three regulators are found at virtually all detected promoters. Moreover, we found that the ability of PigR to specifically associate with promoter regions was dependent upon MglA. We identified a potential mechanism for specificity involving a sequence motif in PigR-regulated promoters, which we have tentatively called the PigR response element (PRE). Finally, although PigR, MglA, and SspA were thought to control the same set of genes, we have been able to discern a difference between the MglA/SspA and PigR regulons. Transcriptomic analyses suggest that PigR only acts as a positive regulator, while MglA appears to act both as a positive regulator in concert with PigR and a negative regulator independently of PigR, with MglA negatively regulating genes in the σ^{32} regulon. Together, our data lead to a model in which the MglA-SspA complex promotes formation of the σ^{70}-containing RNAP holoenzyme and is found at all promoters, providing a point of contact between PigR and RNAP. In this model, PigR interacts with the RNAP-associated MglA-SspA complex at all promoters but only activates transcription from those that contain the PRE.

Introduction

Francisella tularensis is a Gram-negative bacterium and the etiological agent of tularemia. This intracellular pathogen causes disease in small rodents and can be transmitted by a wide variety of insect vectors (Petersen et al., 2009). Humans can become infected by *F. tularensis* in a variety of ways, including exposure to an infected animal or vector, exposure to contaminated food or water, or exposure to aerosolized bacteria (Tärnvik and Chu, 2007). Because the infectious dose is extremely low and because certain strains of *F. tularensis* cause disease with a high incidence of mortality, the organism is considered a potential bioweapon (Dennis et al., 2001). Understanding how these bacteria can infect the host, survive in the host environment, and replicate to continue the infection is thus of great importance and interest.

Many studies and screens have been performed to identify factors critical to the virulence of *F. tularensis*. It has become clear that several transcription factors are critical for pathogenicity. MglA, a stringent starvation protein A (SspA) family member, was the first virulence factor demonstrated to be important for the intracellular survival and replication of *F. tularensis* (Baron and Nano, 1998). In *E. coli*, SspA is one of the most highly produced proteins during the stringent response and has been implicated in mediating the response to cellular stress (Ishihama and Saitoh, 1979; Williams et al., 1994). *F. tularensis* encodes one other SspA family member, named SspA, and both MglA and SspA have been found to be critical for virulence gene expression (Charity et al., 2007). It has been demonstrated that MglA and SspA form a heteromeric complex that interacts with RNA polymerase (RNAP). The MglA-SspA complex interacts with an additional protein called PigR in *F. tularensis* subsp. *holarctica* LVS (also known as FevR in *F. tularensis* subsp. *novicida*), to regulate a common set of genes (Brotcke and Monack, 2008; Charity et al., 2007; 2009). The shared regulon of MglA, SspA, and PigR encompasses approximately 100 genes, including those encoding known virulence factors as well as many whose products are not known to be linked to virulence (Brotcke and Monack, 2008; Brotcke et al., 2006; Charity et al., 2007; 2009). It is notable that MglA, SspA, and PigR

are critical for expression of the genes found on the Francisella Pathogenicity Island (FPI), which encodes a type VI-like secretion system demonstrated to be indispensible for virulence (reviewed in Bröms et al., 2010).

Previous work performed in this laboratory led to a model for virulence gene regulation by MglA, SspA and PigR (Figure 2.1 and Charity et al., 2009). In this model, PigR, a putative DNA-binding protein, binds specifically to a DNA sequence present at the promoters of target genes. DNA-bound PigR functions as a transcription activator by contacting the RNAP-associated MglA-SspA complex at these target promoters, thus stabilizing the binding of RNAP.

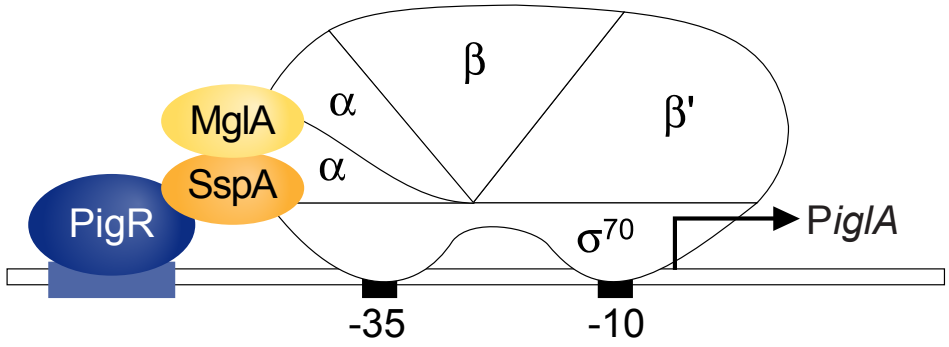

Figure 2.1. A working model for how the MglA-SspA complex and PigR control a common set of genes. PigR, a putative site-specific DNA-binding protein, interacts directly with the RNAP-associated MglA-SspA complex and stabilizes the binding of RNAP at specific target promoters such as the *iglA* promoter found on the FPI.

The model specifies that PigR, like other classical transcription activators, is a site-specific DNA-binding protein that only associates with the promoter regions of MglA/SspA/PigR regulated genes. This model also specifies that PigR interacts with the RNAP-associated MglA-SspA complex at these specific target promoters; the MglA-SspA complex is therefore expected to be present at these target promoters. This model does not address whether the MglA-SspA complex might be found at other promoters whose activity is not regulated by PigR. Furthermore, although the model specifies that PigR interacts with the MglA-SspA complex that is associated with the RNAP holoenzyme during transcription initiation, it does not address whether the MglA-SspA complex is associated with the RNAP core enzyme during transcription elongation.

Using chromatin immunoprecipitation followed by high-throughput DNA sequencing (ChIP-Seq), we show that PigR, MglA, and SspA are present at virtually all detected promoters in *F. tularensis*. We also demonstrate that PigR requires MglA (and thus presumably the MglA-SspA complex) in order to specifically associate with promoters. Finally, we present evidence that the promoters of PigR-regulated genes contain a specific sequence motif. Our findings result in a revised model for the regulation of virulence gene expression in which PigR contacts the RNAP-associated MglA-SspA complex at all promoters. However, PigR may only function as an activator in concert with MglA and SspA at those promoters that contain a specific sequence motif. Transcriptomic analyses provide support for this model, as it was found that PigR, in concert with MglA, only appears to function as a positive regulator. MglA, on the other hand, appears to not only positively regulate a large number of genes in concert with PigR but also negatively regulates the expression of σ^{32}-controlled genes.

Results

Defining promoter regions in *F. tularensis* using ChIP-Seq

In order to address the question of whether PigR specifically associates with target promoters, we first sought to define the locations of all promoters in *F. tularensis*. To do this, we determined the locations of the β′ subunit of RNA polymerase (RNAP) on the chromosome using ChIP-Seq. To facilitate the immunoprecipitation of the β′ subunit of RNAP, we constructed a strain of *F. tularensis* LVS in which the chromosomal copy of the *rpoC* gene (encoding β′) was modified to encode the β′ subunit with a vesicular stomatitis virus-glycoprotein (VSV-G) epitope tag fused to its C-terminus (Figure 2.2A). This results in cells which synthesize the β′ subunit of RNAP with a VSV-G tag at close to native levels (Figure 2.2B). Because the β′ subunit is a core subunit of RNAP, it will be found at both promoter regions and within actively transcribed genes. Thus, enrichment of the β′ subunit within a particular region is not exclusively indicative of promoter location. In order to use the strain encoding the VSV-G tagged β′ protein to specifically identify promoter locations, we performed ChIP-Seq after treatment of the cells with the small molecule rifampicin (rif) to effectively trap RNAP at promoters (Campbell et al., 2001; Herring et al., 2005). By determining the location of the β′ RNAP subunit in cells treated with rif, we identified 526 promoter regions in *F. tularensis* LVS.

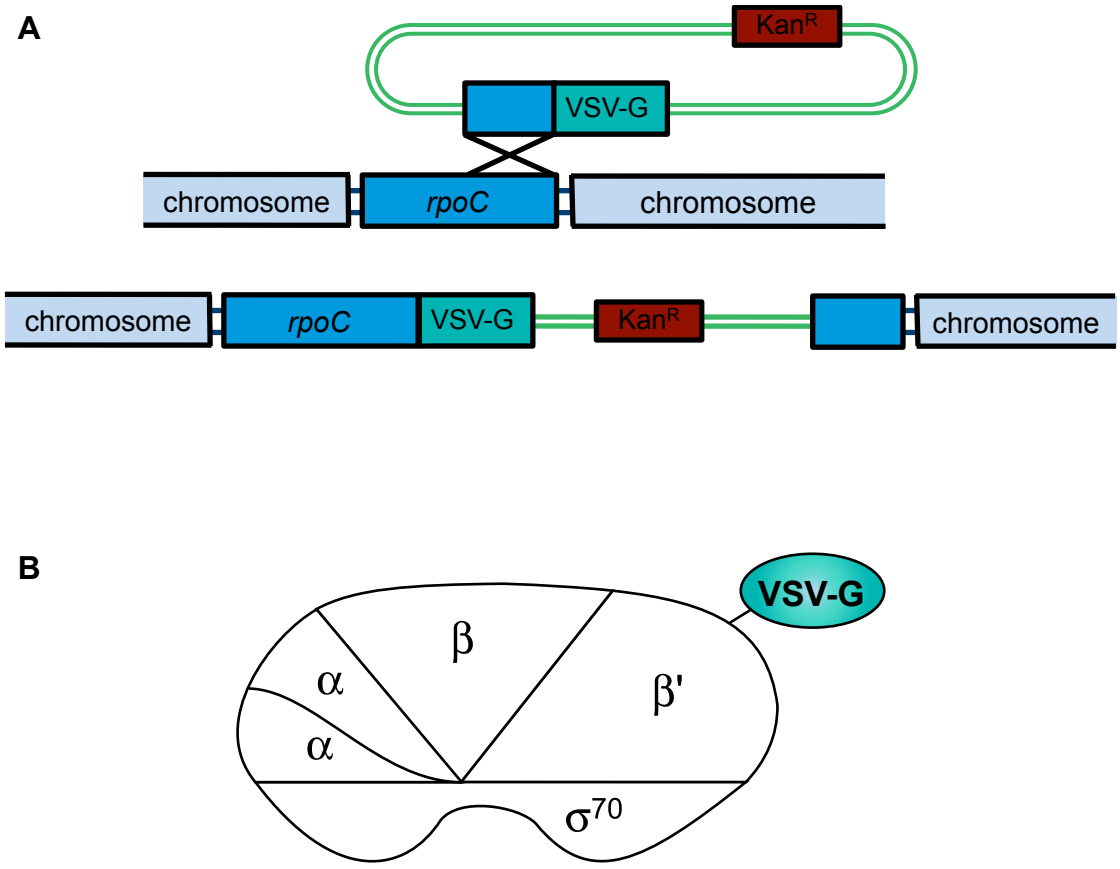

Figure 2.2. Schematic diagram for producing strains with epitope-tagged transcription factors. A. The integration vector contains a fragment of the *rpoC* gene (FTL_1743) followed by DNA specifying a VSV-G epitope tag. A single homologous recombination event produces a strain, LVS β′-V, that synthesizes the VSV-G tagged β′ subunit of RNAP (β′-V) from the native *rpoC* locus. **B.** The VSV-G tagged β′ subunit is incorporated into the RNAP holoenzyme complex.

F. tularensis only encodes two σ factors: σ^{70}, the so-called housekeeping σ factor, and σ^{32}, the so-called heat-shock σ factor (Grall et al., 2009). As a complementary approach to identify promoters in *F. tularensis*, and to determine which promoters are σ^{70}-dependent and which are controlled by σ^{32}, we performed ChIP-Seq on both σ factors. We used the strategy used above to modify the chromosomal *rpoD* and *rpoH* genes, encoding σ^{70} and σ^{32} respectively, to encode the corresponding RNAP subunit with a VSV-G epitope tag fused to its C-terminus. We also chose to similarly modify *rho*, the gene encoding the transcription termination factor Rho, as an example of a protein that associates with RNAP at non-promoter regions, and *oxyR*, which encodes the oxidative stress transcription regulator OxyR, as an example of a site-specific DNA-binding protein and transcription factor which exerts its regulatory effects largely at promoter regions. The regions identified by determining the location of σ^{70} overlapped the majority of the promoters that were identified by determining the location of the β′ RNAP subunit in cells grown in the presence of rif. However, under our growth conditions, only four regions were identified as enriched for σ^{32}. By defining a promoter as a region with significant enrichment of either σ factor and/or the β′ subunit of RNAP in the presence of rif, we identified 582 promoter regions in *F. tularensis* LVS (Table 2.1).

Table 2.1. Promoter regions identified by ChIP-Seq

Factor	Peaks
β′ + rif	526
σ^{70}	333
σ^{32}	4
Total promoters	582

Figure 2.3 contains an example of a section of the genome with regions identified as enriched after ChIP-Seq of β′, β′+rif, σ^{70} and σ^{32}. The β′ subunit of RNAP is found both at promoter regions and within actively transcribed genes. Determining the location of the β′ subunit of RNAP after rif treatment specifically identifies promoter regions, which correlate well to regions of σ factor enrichment. These data suggest that the region between FTL_1409 and FTL_1410 contains a σ^{70}-dependent promoter, while the region between FTL_1410 and FTL_1411 contains a σ^{32}-dependent promoter (Figure 2.3).

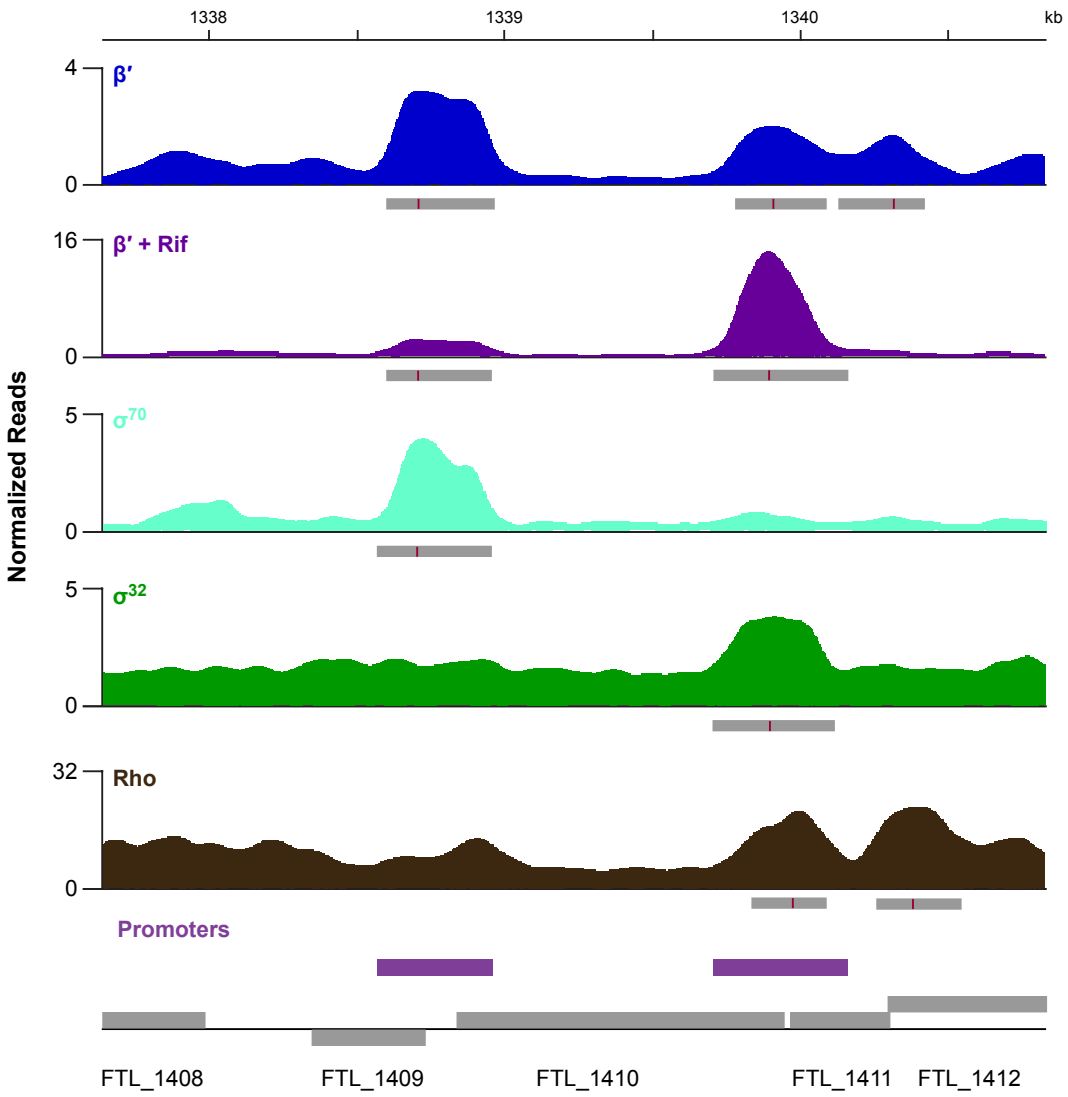

Figure 2.3. Putative promoter regions are identified using ChIP-Seq. A representative section of the chromosome is depicted. Genomic position is indicated on the X-axis by *F. tularensis* LVS locus number; gray boxes represent genes, which are above the black line if they are encoded on the plus strand and below the black line if they are encoded on the minus strand. A representative track illustrating the density of normalized mapped sequencing reads is depicted on the Y-axis after ChIP-Seq of each factor, β′ (blue), β′+ rif (purple), σ^{70} (aqua), σ^{32} (green), and Rho (brown). Areas of significantly enriched reads, referred to here as peaks, are indicated by the horizontal gray boxes below the read density plot and sites of maximum enrichment are indicated by the red lines. Putative promoter regions, which are areas with significant enrichment of β′ + rif, σ^{32}, or σ^{70} association with the chromosome, are indicated by the purple boxes above the gene annotations.

MglA, SspA, and PigR are found at essentially all promoters in *F. tularensis*

Having defined the location of putative promoter regions across the genome, we next determined at which promoters PigR, MglA, and SspA were located. We utilized a tagging strategy similar to the one used with the RNAP subunits, which allowed us to modify the chromosomal copy of each gene to encode a protein containing a C-terminal epitope tag. Cells from the resulting strains were used for ChIP-Seq to determine the chromosomal locations of PigR, MglA, and SspA.

These data revealed that PigR, MglA, and SspA are located specifically at promoters (i.e. they are not found to be enriched throughout the gene similarly to Rho), and at not only candidate MglA/SspA/PigR regulated promoters, but also at the majority of promoters throughout the genome. This is illustrated in Figure 2.4, which depicts the data from the *iglA*, *proS* and FTL_0651 promoter regions. Expression of the *iglA* gene is dependent on the presence of MglA, SspA, and PigR, whereas the expression of *proS* and FTL_0651 are not. In contrast, we did not identify enrichment of OxyR at either promoter, indicating that we are not simply immunoprecipitating all promoters with every transcription factor. The identification of PigR at essentially all promoters suggests that PigR is not a sequence-specific DNA-binding protein that is only found at specific target promoters.

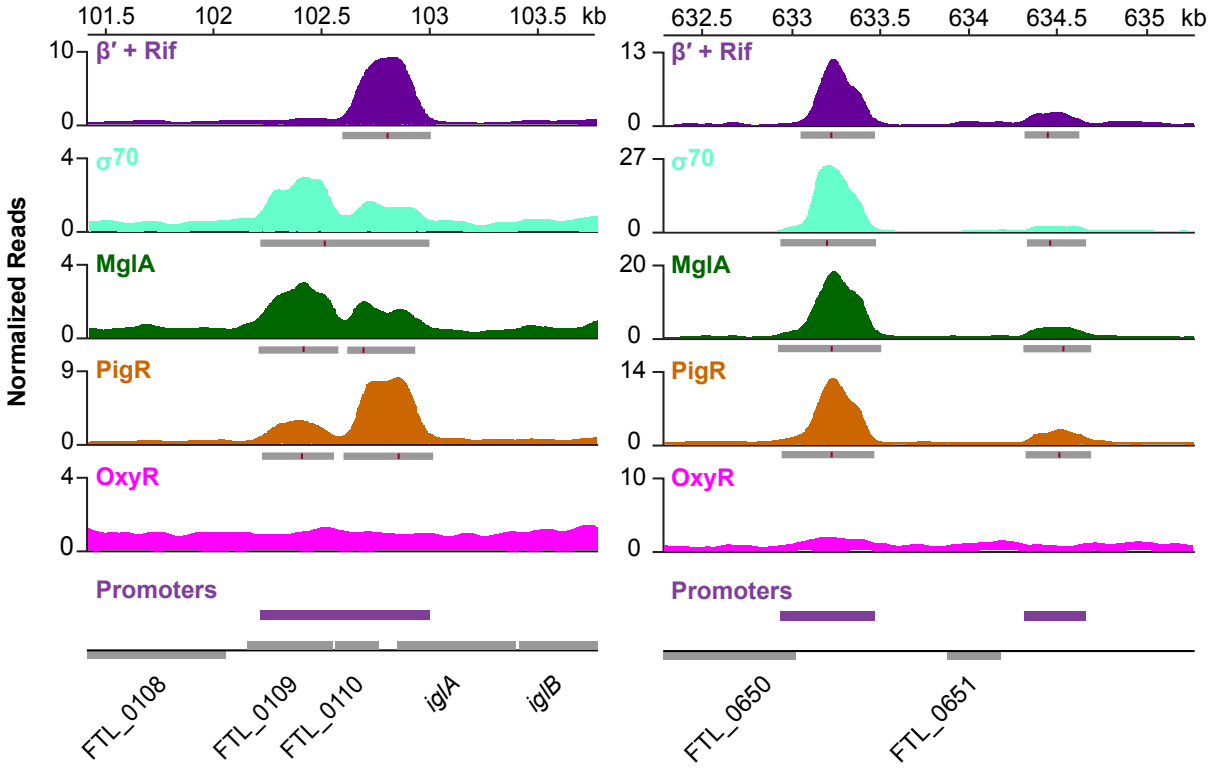

Figure 2.4. PigR and the MglA-SspA complex co-localize at the promoters of target and non-target genes. Representative sections of the chromosome are depicted, including promoter regions for both a target gene (FTL_0111, FPI virulence gene *iglA*, left) and non-target regions (FTL_0650, *proS,* encoding a tRNA synthase and FTL_0651, encoding a transposase). Genomic position is indicated on the X-axis by *F. tularensis* LVS locus number; gray boxes represent genes, which are above the black line if they are encoded on the plus strand and below the black line if they are encoded on the minus strand. A representative track illustrating the density of normalized mapped sequencing reads is depicted on the Y-axis after ChIP-Seq of each factor, β′+ rif (purple), σ^{70} (aqua), MglA (green), PigR (orange), and OxyR (pink). Areas of significantly enriched reads, referred to here as peaks, are indicated by the horizontal gray boxes below the read density plot and sites of maximum enrichment are indicated by the red lines. Putative promoter regions, which are areas with significant enrichment of β′ + rif, σ^{32}, or σ^{70} association with the chromosome, are indicated by the purple boxes above the gene annotations. The concordance between enrichment of the RNAP β′ subunit, σ^{70}, MglA, and PigR is in contrast to the transcription factor OxyR.

The concordance among the localization of MglA, SspA, and PigR is further demonstrated in Figure 2.5. The Venn diagram in Figure 2.5 represents the 98% of promoter regions identified by ChIP-Seq of σ^{70} at which at least one of the three factors, MglA, SspA, or PigR, are found. The percentages in Figure 2.5 indicate the concordance of localization between MglA, SspA, and PigR at σ^{70}-associated promoters. All three factors, MglA, SspA, and PigR, are found at the majority of promoters identified by detection of σ^{70}.

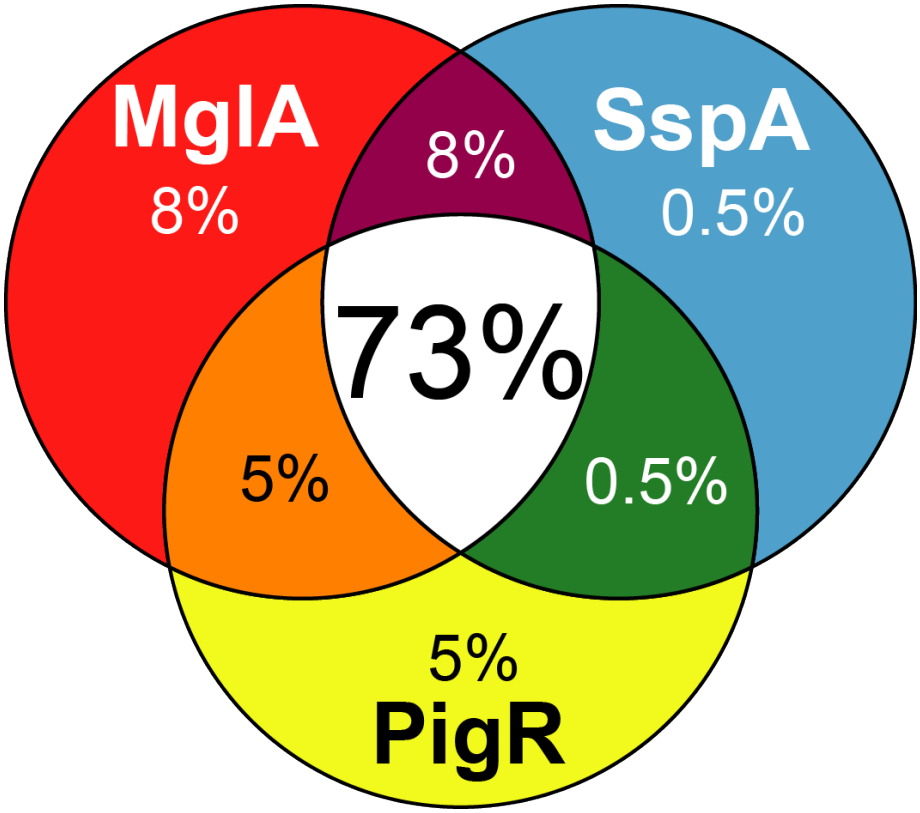

Figure 2.5. The MglA-SspA complex and PigR are present at the majority of promoters. The overlap between MglA, SspA, and PigR ChIP-Seq peaks at promoter regions identified by ChIP-Seq of σ^{70} is depicted (Venn diagram is not to scale). Diagram represents 326 of the 333 (i.e., 98%) ChIP-Seq peaks identified by σ^{70}.

PigR requires MglA in order to specifically associate with promoters

Since PigR was found together with MglA-SspA at the majority of promoters, and given that PigR may be a DNA-binding protein, it was unclear if PigR is found at all promoter regions regardless of the presence of the MglA-SspA complex, potentially binding a specific DNA sequence found at all promoters, or if PigR requires the presence of the MglA-SspA complex in order to associate with promoter regions. To determine if PigR requires the MglA-SspA complex to associate with promoter regions, we performed ChIP-Seq on PigR in the presence and absence of MglA. In particular, we performed ChIP-Seq with cells of the Δ*pigR* mutant strain and cells of the Δ*mglA* Δ*pigR* mutant strain in which we expressed the *pigR* gene modified to encode the PigR protein with a C-terminal VSV-G tag (*pigR-V*) from a plasmid under the control of a heterologous promoter. The protein encoded by *pigR-V* is functional, as it has been previously demonstrated to complement the Δ*pigR* mutant strain with respect to *iglA* transcript abundance (Charity et al., 2009). It was necessary to drive the expression of *pigR-V* from a heterologous promoter in these experiments because expression of the native *pigR* gene is dependent upon MglA (and SspA) (Brotcke et al., 2006; Charity et al., 2007). We assessed the abundance of the PigR-V protein in cells of the Δ*pigR* mutant and the Δ*mglA* Δ*pigR* mutant strains. While there was less PigR-V produced in the Δ*mglA* Δ*pigR* mutant strain, protein levels were largely comparable (Figure 2.6).

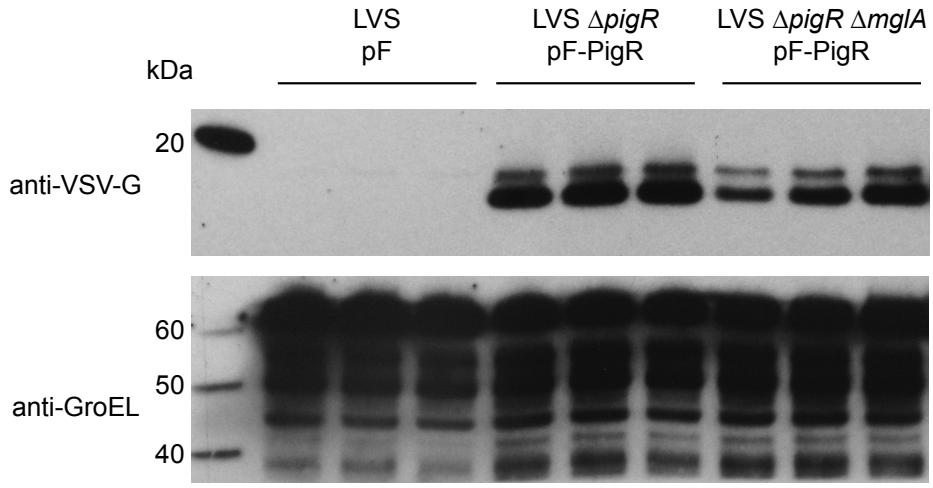

Figure 2.6. The abundance of ectopically produced PigR-V is similar in cells of the Δ*pigR* mutant strain and cells of the Δ*mglA* Δ*pigR* mutant strain. A Western blot analysis of the amount of ectopically produced PigR-V protein. Protein from equivalent amounts of cells from triplicate cultures of (lanes 1-3) LVS cells with the pF plasmid (an empty vector control), (lanes 4-6) LVS Δ*pigR* cells with the pF-PigR-V plasmid, and (lanes 7-9) LVS Δ*pigR* Δ*mglA* cells with the pF-PigR-V plasmid were electrophoresed on a 4-12% Bis-Tris NuPAGE gel and analyzed by Western blotting. Upper panel, immunoblot probed with anti-VSV-G. PigR-V is found in cells of the Δ*pigR* mutant and in cells of the Δ*mglA* Δ*pigR* mutant at comparable levels. Lower panel, immunoblot probed with anti-GroEL. Detection of GroEL (~60 kDa) and the degradation products of GroEL (<60 kDa) serve as a loading control.

Comparison of the ChIP-Seq results obtained with ectopically produced PigR with those obtained with native PigR revealed that ectopic production of PigR does not significantly alter the location of this protein (Figure 2.7). Comparison of the ChIP-Seq results obtained with ectopically produced PigR in the presence and absence of MglA revealed a striking difference in location; we found no specific enrichment of PigR at promoter regions in the absence of MglA (Figure 2.7). This indicates that MglA, and by inference the MglA-SspA complex, is required for PigR to specifically associate with promoter regions.

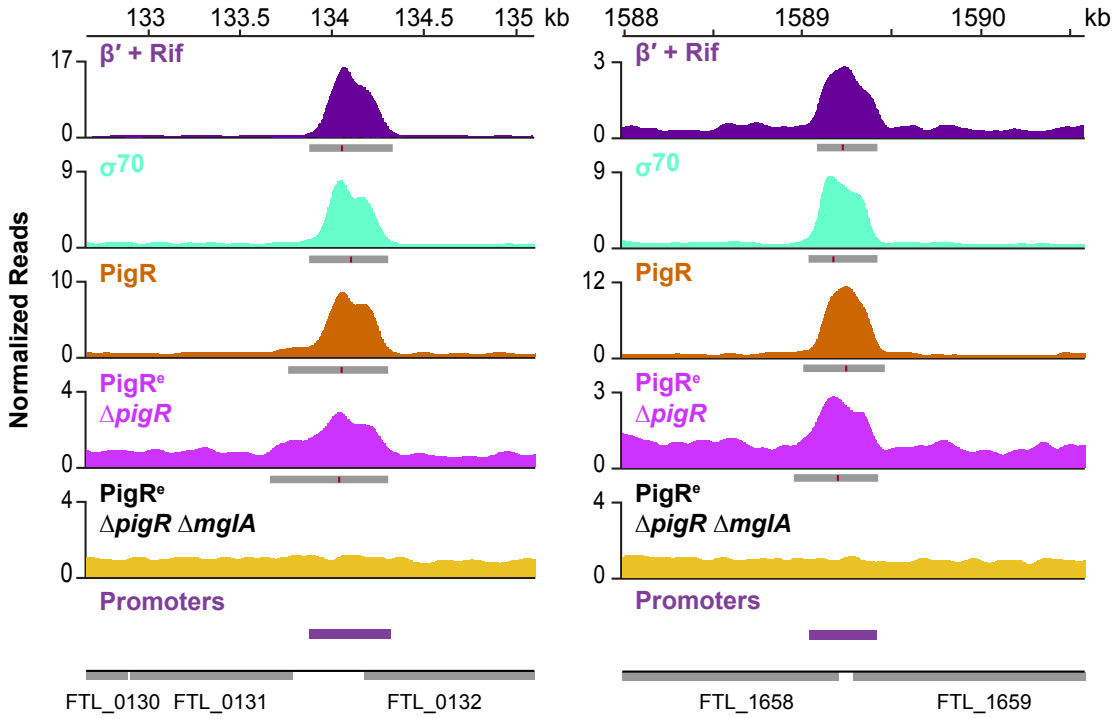

Figure 2.7. PigR requires MglA to specifically associate with promoter regions. Representative sections of the chromosome are depicted, including promoter regions for both a target gene (*ilvE*, FTL_0131, left) and non-target regions (FTL_1658, *fimV,* encoding a Type IV pilus assembly protein). Genomic position is indicated on the X-axis by *F. tularensis* LVS locus number; gray boxes represent genes, which are above the black line if they are encoded on the plus strand and below the black line if they are encoded on the minus strand. A representative track illustrating the density of normalized mapped sequencing reads is depicted on the Y-axis after ChIP-Seq of each factor, β′+ rif (purple), σ^{70} (aqua), PigR (orange), ectopically expressed PigR from Δ*pigR* cells (pink), and ectopically expressed PigR from Δ*pigR* Δ*mglA* cells (yellow). Areas of significantly enriched reads, referred to here as peaks, are indicated by the horizontal gray boxes below the read density plot and sites of maximum enrichment are indicated by the red lines. Putative promoter regions, which are areas with significant enrichment of β′ + rif, σ^{32}, or σ^{70} association with the chromosome, are indicated by the purple boxes above the gene annotations. This region illustrates that enrichment of ectopically expressed PigR in a Δ*pigR* strain is similar to chromosomally encoded PigR. In comparison, there is a marked lack of promoter-specific enrichment of ectopically expressed PigR in a Δ*pigR* Δ*mglA* strain.

Cells lacking MglA or PigR have different growth rates

Because MglA, SspA, and PigR appear to regulate the expression of the same set of genes, and because these three factors are present at virtually all promoters, we were interested in determining if cells lacking either MglA or PigR differed at all phenotypically. We observed, consistent with previously reported findings, that cells of the LVS Δ*mglA* mutant strain exhibit a slow growth phenotype (Figure 2.8; Charity et al., 2007; Honn et al., 2012). However, we found that a strain with a deletion of *pigR* does not have a significantly altered growth rate (Figure 2.8). We found the doubling times of cells of the wild-type strain, cells of the Δ*pigR* strain, and cells of the Δ*mglA* strain during log phase to be approximately 159, 156, and 203 minutes, respectively. This is a surprising result, if the only functions of PigR and MglA are to regulate exactly the same set of genes. These data indicate that there are differences between PigR and MglA, and MglA may be regulating something important for maintaining growth rate.

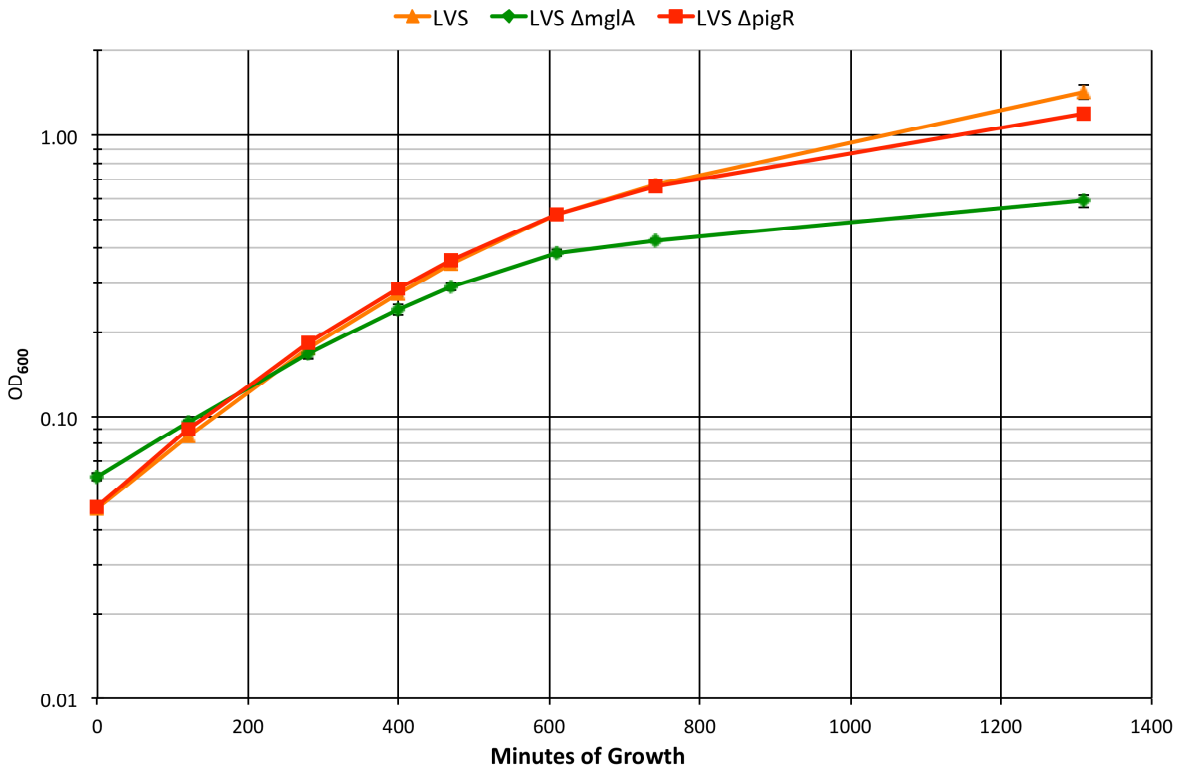

Figure 2.8. Cells of the Δ*pigR* mutant strain grow similarly to wild-type cells whereas cells of the Δ*mglA* mutant strain grow more slowly than wild-type cells. Each dot represents the average OD_{600} value of three cultures at the indicated time point, grown in supplemented Mueller-Hinton broth, with standard deviations marked.

PigR and MglA positively regulate the expression of the same set of genes

Using ChIP-Seq we found MglA, SspA, and PigR present at virtually all promoter regions. Additionally, previous studies found that in *F. tularensis* there is significant overlap in the genes regulated by MglA, SspA, and PigR (Brotcke and Monack, 2008; Charity et al., 2009). Yet the loss of MglA results in a slow growth phenotype while the loss of PigR does not. We reasoned that this might be the result of differences between the genes that MglA and PigR regulate.

We asked if we could detect any differences between the regulons of MglA and PigR using an accurate and sensitive method. We utilized Nanostring to examine 100 transcripts, 19 from genes previously shown to be positively regulated by MglA/SspA, 21 genes reported as negatively regulated by MglA/SspA, and 60 genes which have not been previously reported to be MglA/SspA regulated (Brotcke et al., 2006; Charity et al., 2007; Geiss et al., 2008). We compared the abundance of the described transcripts in wild-type cells to that in cells lacking either MglA or PigR. The results confirmed the positive regulation of 13 genes by both MglA and PigR (Table 2.2). Of the previously reported positively controlled genes, transcript levels of two were too variable to be significant and transcript levels of four others were found to be significantly regulated by either MglA or PigR, but did not reach the two-fold cutoff for the other factor.

Table 2.2. Genes positively regulated by MglA and/or PigR

Locus	Gene Name	Fold Change ΔmglA	Fold Change ΔpigR	Gene Product	MglA/SspA regulated[1]
FTL_0449	*pigR*	-27.6	NA**	PigR transcription factor	+
FTL_0111	*iglA*	-26.9	-23.9	Intracellular growth locus, subunit A	+
FTL_0026	-	-25.5	-19.1	3-hydroxyisobutyrate dehydrogenase	+
FTL_0113	*iglC*	-14.1	-10.6	Intracellular growth locus, subunit C	+
FTL_1219	-	-10.6	-4.9	Hypothetical protein	+
FTL_0126	*pdpA*	-9.0	-14.7	Hypothetical protein	+
FTL_1218	-	-7.7	-5.9	Hypothetical protein	+
FTL_0116	*pdpC*	-5.8	-7.9	Hypothetical protein	+
FTL_0879	*bla*	-5.0	-4.5	beta-lactamase	+
FTL_1790	*ampG*	-4.7	-2.6	major facilitator superfamily protein	+
FTL_0125	*pdpB*	-4.4	-8.5	Hypothetical protein	+
FTL_0500	*speE*	-4.2	-2.0	Spermidine synthase	+
FTL_0491	-	-4.1	-4.3	Outer membrane lipoprotein	+
FTL_0617	*bfr*	-2.5	-1.3*	bacterioferritin	+
FTL_0477	*gcvT*	-2.3	-1.2	glycine cleavage system aminomethyltransferase T	+
FTL_0068	*gmhA*	-2.3	-1.1*	phosphoheptose isomerase	ND
FTL_1731	-	-2.2	-1.0*	licB-like transmembrane protein	ND
FTL_0133	*feoB*	-2.2	-1.3	ferrous iron transport protein	ND
FTL_0317	-	-2.1	1.1	Hypothetical protein	ND
FTL_1727	*ampD*	-2.1	-1.6	N-acetyl-anhydromuranmyl-L-alanine amidase	ND
FTL_1756	*glpA*	-2.0	-1.6	anaerobic glycerol-3-phosphate dehydrogenase	ND
FTL_0646	-	-1.9	-3.7	Hypothetical protein	-
FTL_1678	-	-1.8	-3.0	conserved membrane hypothetical protein	+
FTL_0131	*ilvE*	-1.5	-2.0	branched-chain amino acid aminotransferase protein (class IV)	+
FTL_0388	-	-1.3	-2.0	cation transporter	ND

[1] "+", gene previously described as positively regulated; "-", gene previously described as negatively regulated; "ND", gene not previously described as MglA/SspA regulated.

* Not significant, T-test between deletion strain and wild-type LVS results in a p-value > 0.05.

** Not applicable, no detectable *pigR* transcript in the Δ*pigR* mutant strain.

A significant difference between previous reports and these results is the presence of six additional genes, not previously reported as MglA-regulated, which have transcripts reduced more than 2-fold in the Δ*mglA* strain. While MglA is thought to regulate the expression of over 100 genes, none of these genes have previously been reported to be under the control of MglA. They are all closer to the lower limit of the significance threshold than any of the previously reported transcripts. This suggests, not surprisingly, both that the Nanostring assay is likely more sensitive than the previous microarray analyses and, if this subset of genes is representative of the rest of the genome, that MglA could be affecting the expression of many more genes than previously thought.

We found that the PigR regulon differed significantly from the MglA regulon in one principal way. Specifically, no transcripts were identified as being negatively regulated by PigR, in contrast to the 15 transcripts negatively regulated by MglA (Table 2.3). This indicates PigR appears to positively regulate the expression of most of the genes that are positively regulated by MglA but does not influence the expression of those genes that are negatively regulated by MglA.

Table 2.3. Genes negatively regulated by MglA

Locus	Gene Name	Fold Change ΔmglA	Fold Change ΔpigR	Gene Product	MglA/SspA regulated[1]	Putative σ^{32}-regulon[2]
FTL_1265	*folK*	3.6	-1.5*	2-amino-4-hydroxy-6-hydroxymethyldihydropteridine pyrophosphokinase/ dihydropteroate synthase	ND	+
FTL_1600	*ansB*	2.9	-1.3*	Periplasmic L-asparaginase II precursor	-	+
FTL_1103	-	2.7	-1.2*	DNA helicase, pseudogene	-	-
FTL_1273	*bioF*	2.7	-1.2	8-amino-7-oxononanoate synthase	-	-
FTL_1050	*rpoD*	2.7	-1.1*	RNA polymerase sigma-70 factor	-	-
FTL_0528	-	2.7	1.1*	Type III restriction enzyme	-	-
FTL_0450	*psd*	2.6	-1.0*	phosphatidylserine decarboxylase	-	-
FTL_0056	-	2.4	-1.4*	NADH dehydrogenase	-	-
FTL_1190	*grpE*	2.3	-2.8*	heat shock protein GrpE	-	+
FTL_1715	*groES*	2.3	-2.7*	co-chaperonin GroES	ND	+
FTL_1421	-	2.2	1.0*	haloacid dehalogenase-like hydrolase family protein	-	-
FTL_1948	-	2.2	-1.3	Major facilitator superfamily (MFS) transport protein	-	-
FTL_1449	*rnd*	2.0	1.0*	ribonuclease D	-	-
FTL_1266	-	2.0	-1.5	lipase/esterase	ND	+
FTL_0660	*feoA*	2.0	1.2	ferrous iron transport protein A	ND	-

[1] "+", gene previously described as positively regulated; "-", gene previously described as negatively regulated; "ND", gene not previously described as MglA/SspA regulated.

[2]"+", gene is part of the putative σ^{32} regulon; "-", gene is not part of the putative σ^{32} regulon; as determined by this study and Grall et al., 2009.

* Not significant, T-test between deletion strain and wild-type LVS results in a p-value > 0.05.

The putative σ^{32} regulon is comprised of genes whose transcripts are reported to be significantly upregulated upon heat-shock or overexpression of σ^{32} (Grall et al., 2009), and/or have translational start sites within either 100 bp upstream or 1 kb downstream from the maximal point of a σ^{32} ChIP-Seq peak. Many of the genes negatively regulated by MglA belong to this putative σ^{32} regulon. Direct comparison between cells lacking MglA and cells lacking PigR highlights that a significant difference between the two strains is the increased abundance of σ^{32}-dependent transcripts in cells lacking MglA (Table 2.4). Higher levels of σ^{32}-regulated transcripts in cells lacking MglA are consistent with the negative regulation of σ^{32}-regulated genes by MglA but not by PigR.

Table 2.4. Genes negatively regulated by MglA in comparison to PigR

Locus	Gene Name	Fold Change ΔpigR	Gene Product	MglA/SspA regulated[1]	Putative σ32-regulon[2]
FTL_1190	*grpE*	-6.5	heat shock protein GrpE	-	+
FTL_1715	*groES*	-6.3	co-chaperonin GroES	ND	+
FTL_1265	*folK*	-5.3	2-amino-4-hydroxy-6-hydroxymethyldihydropteridine pyrophosphokinase/dihydropteroate synthase	ND	+
FTL_1600	*ansB*	-3.8	Periplasmic L-asparaginase II precursor	-	+
FTL_0281	*hsp40*	-3.7	heat shock protein, hsp40	ND	+
FTL_1103	-	-3.3	DNA helicase, pseudogene	-	-
FTL_0056	-	-3.3	NADH dehydrogenase	-	-
FTL_1273	*bioF*	-3.2	8-amino-7-oxononanoate synthase	-	-
FTL_1266	-	-3.1	lipase/esterase	ND	+
FTL_1050	*rpoD*	-2.8	RNA polymerase sigma-70 factor	-	-
FTL_1948	-	-2.8	Major facilitator superfamily (MFS) transport protein	-	-
FTL_1372	-	-2.8	lipoprotein	ND	-
FTL_0450	*psd*	-2.6	phosphatidylserine decarboxylase	-	-
FTL_0528	-	-2.4	Type III restriction enzyme	-	-
FTL_0178	*yidC*	-2.2	inner-membrane protein	-	-
FTL_1421	-	-2.2	haloacid dehalogenase-like hydrolase family protein	-	-
FTL_0892	*clpP*	-2.1	ATP-dependent Clp protease proteolytic subunit	ND	+
FTL_0283	-	-2.1	aromatic amino acid HAAP transporter	ND	-

[1] "+", gene previously described as positively regulated; "-", gene previously described as negatively regulated; "ND", gene not previously described as MglA/SspA regulated.

[2]"+", gene is part of the putative σ^{32} regulon; "-", gene is not part of the putative σ^{32} regulon; as determined by this study and Grall et al., 2009.

Our findings suggest that MglA and PigR differ in their functions, such that PigR works in concert with MglA (and, by inference, SspA) to positively control gene expression, while MglA (and SspA) also negatively effects the expression of certain genes. Specifically, one function of the MglA-SspA complex may be to repress the expression of σ^{32}-regulated genes.

A specific sequence motif is found at the promoters of MglA/SspA/PigR-regulated genes

While PigR is present at virtually all identified promoters, it appears to only promote the expression of a fraction of the corresponding genes. We hypothesized that while PigR is present at all promoters, PigR may only promote gene expression at specific promoters upon recognition of a specific sequence element. Using MEME, we searched for a motif in the promoter regions of genes that matched certain criteria (Bailey and Elkan, 1994). Specifically, genes must have been previously reported as PigR regulated in LVS (Charity et al., 2009) or demonstrated to be PigR-regulated in our Nanostring studies and have a PigR-promoter peak (as determined by our ChIP-Seq studies) upstream from the translation start site. Eleven genes fit our criteria and a 7 bp motif was found to be present in all 11 of the promoter regions analyzed (Figure 2.9A). A logo representing the 7 bp motif can be seen in Figure 2.9C. The transcription start site of 5 PigR-regulated genes has been identified and this motif maps to the same site relative to the transcription start site in all these regions, 6-8 bp upstream from the predicted -35 element (Figure 2.9B). We compared this region in PigR-regulated promoters to the same region in promoters mapped by Zaide et al., none of which appear to be regulated by PigR; the 7 bp motif was not found at these non-PigR regulated promoters (Zaide et al., 2011) (Figure 2.9B). This suggests that the motif is specific to PigR-regulated promoters, raising the possibility that PigR may bind directly to this site to activate transcription from regulated promoters.

A

Sequence name	Start	P-value		PigR motif	
FTL_1218	194	3.76e-05	TTGTAACCCG	TGCTGTA	TATCCTATAT
FTL_0449	137	3.76e-05	CTGAAATAAT	TGCTGTA	GAAAAGGTTG
FTL_0207	187	3.76e-05	CTGCAATATA	TGCTGTA	AAAGATTTTA
FTL_0126	270	1.23e-04	CAGACCAATT	AGCTGTA	AACATATAGT
FTL_0111	116	1.23e-04	GTTTAAATTT	AGCTGTA	TAAACATTGT
FTL_1790	105	1.69e-04	TTATTTTTTT	TGCTCTA	TTCTTTATTT
FTL_0491	150	1.69e-04	TAAAAAAATA	TGCCGGA	GCATTCTCTT
FTL_1219	90	2.77e-04	AAAAATATTT	TGCTATA	AAATTTATTT
FTL_0026	167	3.43e-04	AAATAGTATT	TGCAGTA	AAAAAGTGTT
FTL_1678	73	4.00e-04	TATTATAAAT	TGCTAGA	TATTATTAAT
FTL_0131	115	4.70e-04	ATCATTAATA	CGCTATA	ATCTAGGTTT

B

Locus		PigR motif		-35		-10		TSS
FTL_0026	ATT	TGCAGTA	AAAAAG	TGTTAA	TCAGTTTTGTGAGGTAC	TAAAAT	TAAATAT	A
FTL_0111	TTT	AGCTGTA	TAAACA	TTGTGT	TATTGGCGTTATTAAGG	TAACTT	GCTTAT	A
FTL_0126	ATT	AGCTGTA	AACATA	TAGTCA	ATAGATAAGTGCTTATG	TAAAAT	GACTTT	A
FTL_1219	TTT	TGCTATA	AAATTTAT	TTCAAA	ATGAAATTGATTAATGCA	TACTTA	CTGATA	A
FTL_1790	TTT	TGCTCTA	TTCTTTA	TTTACT	TTCTTAGTTATTTCAAC	TAGCAT	TTTATCT	A
FTL_0617	TTTTTATTAAAATATCTAGG			TTGAAT	TCTTAGATATTTTGATA	TATAAT	TAGATACTA	A
FTL_0508	GATACAATATCTATTAATAT			ATTTAA	GCTCAAGAAACTGATA	TAATTT	AGCCACTTT	A
FTL_0199	TCTGATATAGTTAATTTACA			AAGGCT	AAATACTATTATAAAA	TCTAAA	CATTAAA	C
FTL_1138	GCTTAATTTTTGCAAAATTA			TTGCAA	AAATGATTAAAAAATAA	TATTAT	TGACTCG	A
FTL_1715	GACAAAAAAACAAAAAAATT			TCTTGA	AAATTTTTTTTTTGAC	TCAATA	TCTAGACT	T

C

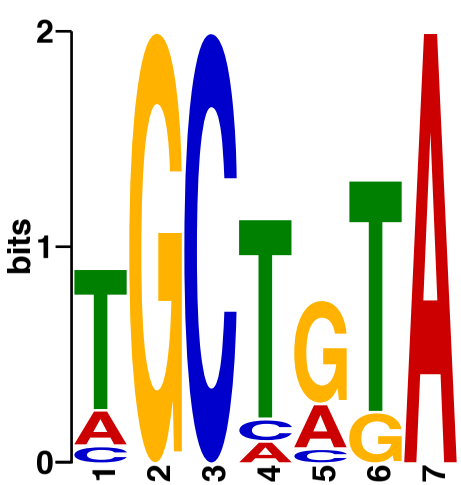

Figure 2.9. **A motif upstream of the -35 element is conserved in PigR-regulated promoters. A.** Alignment generated by MEME of a motif found in PigR-regulated promoters. **B.** Alignment of the promoter regions of four PigR-regulated promoters and five non-PigR-regulated promoters with mapped transcription start sites (transcription start sites for FTL_0026 and FTL_1790 were identified using RNA-Seq [K. Levasseur, B.E. Nickels, and S.L. Dove, unpublished], whereas those for FTL_0111, FTL_0126 and FTL_1219 were mapped by primer extension analyses [J. Charity, and S.L. Dove, unpublished], and those for FTL_0617, FTL_0508, FTL_0199, FTL_1138, and FTL_1715 were determined by Zaide et al. [Zaide et al., 2011]).

Discussion

We have shown that MglA, SspA, and PigR are found at virtually all promoter regions in LVS, and that the association of PigR with promoters is dependent upon the presence of MglA. Furthermore, we have identified a sequence motif present upstream of the -35 element of promoters that may make them responsive to PigR. In addition, we identified differences between the functions of PigR and MglA. Specifically, MglA appears to function as both a positive and negative regulator and provides a function that maintains the normal growth rate of the cell. PigR appears to only act as a positive regulator of gene expression, only influencing the expression of those genes that are positively regulated by MglA and SspA.

Finding PigR, MglA, and SspA at virtually all promoters suggests that these proteins are associated with the RNAP holoenzyme at all promoters. There is at least one other transcription regulator studied in other organisms that is also found at virtually all promoters. CarD is an essential transcription factor in *Mycobacterium tuberculosis* which directly interacts with RNAP to simulate transcription initiation (Srivastava et al., 2013). While PigR, MglA, and SspA are not essential transcription factors and are not thought to be homologous to CarD, it is possible that they function in an analogous manner.

Consistent with the idea that a heteromer of MglA and SspA interacts with the σ^{70}-containing RNAP holoenzyme, σ^{70} together with the core subunits of RNAP and SspA were found to co-purify with MglA in LVS in stoichiometric amounts (Charity et al., 2007). There is also evidence that the SspA homodimer in *E. coli* promotes gene expression from specific promoters when associated with a site-specific DNA-binding protein, Lpa, and σ^{70}-containing RNAP holoenzyme, but not the RNAP core enzyme or RNAP holoenzyme containing the alternate sigma factor, σ^{S} (Hansen et al., 2003).

Both MglA and SspA lack any obvious DNA-binding motifs, and the crystal structure of the SspA homodimer from *Yersina pestis* lacks any obvious DNA-binding determinant (Hansen et al., 2005). Additionally, the *E. coli* SspA homolog was found to activate transcription from the

bacteriophage P1 late promoter P*s*, but not bind to the promoter DNA (Hansen et al., 2003). Thus, there is no indication that the MglA-SspA complex would necessarily bind DNA directly to affect transcription.

PigR requires the MglA-SspA complex in order to specifically localize to promoter regions. We infer from this that interaction between PigR and the RNAP-associated MglA-SspA complex directs PigR specifically to promoter regions. These findings do not rule out the possibility that PigR may also function through interaction with the DNA.

We have identified a sequence motif present upstream of the -35 element of promoters that are positively regulated by PigR (and the MglA-SspA complex). This motif is found 6-8 bp from the predicted -35 element in PigR-responsive promoters. It should be noted that the distance between the -35 element and this motif may actually be a specific number of bp (i.e., either 6 or 7 or 8 bp), and that the variation in location relative to the -35 element could reflect a misannotation of the -35 element.

Because the conserved motif in PigR-regulated promoters has the potential to confer responsiveness to PigR, we tentatively name it the PigR response element (PRE). It should be stressed that further experimentation is required to determine whether or not this motif truly represents a PRE. Nevertheless, the identification of a motif associated with PigR-regulated promoters suggests that PigR may specifically recognize this motif and function to activate those promoters containing this motif by making simultaneous contacts with the DNA and with the RNAP-associated MglA-SspA complex. The fact that PigR is present at many promoters that are not positively regulated by PigR raises the possibility that at most promoter regions, PigR may bind the DNA in a non-sequence specific manner or may not bind the DNA at all.

Although PigR contains a putative helix-turn-helix motif, PigR has yet to be shown to be capable of binding DNA. However, the PRE need not be bound by PigR directly. Association of PigR with the MglA-SspA-RNAP complex may allow contact between the PRE and part of RNAP, such as the α subunit C-terminal domain (αCTD), or even the MglA-SspA complex,

although there is a lack of evidence to suggest that the MglA-SspA complex could bind DNA. It is also possible that PigR might exert effects at some promoters that lack a PRE by contacting the DNA in a non-sequence specific fashion. Indeed, although the αCTD has been shown to bind specifically to the DNA at so-called UP-elements, non-sequence specific binding of the αCTD to the DNA has been shown to influence the activity of several promoters (Ross and Gourse, 2005). It will therefore be important to determine whether all PigR-controlled promoters contain a recognizable PRE.

While PigR was previously thought to control the exact same set of genes as MglA and SspA (Brotcke and Monack, 2008; Charity et al., 2009), we found an important difference between the PigR and MglA/SspA regulons. In particular, using Nanostring, which is a sensitive method of detecting and quantifying RNA transcript abundance, we found that PigR positively regulates the expression of genes that are positively regulated by MglA/SspA but does not negatively regulate the expression of those genes that are negatively regulated by MglA/SspA.

Intriguingly, many of the genes that have been found to be negatively regulated by MglA appear to be driven by σ^{32}-dependent promoters. We offer a possible explanation for this finding. We hypothesize that the MglA-SspA complex promotes formation of the σ^{70}-containing RNAP holoenzyme but not the σ^{32}-containing holoenzyme. This would be consistent with (i) our transcriptomic data demonstrating that σ^{32}-regulated transcripts are more abundant in the absence of MglA (Tables 2.2 and 2.3), with (ii) the finding that MglA co-purifies with σ^{70} and with (iii) the idea that other members of the SspA family of proteins appear to specifically associate with σ^{70}-containing RNAP holoenzyme (De Reuse and Taha, 1997; Hansen et al., 2003; Yin et al., 2013). In the absence of MglA a smaller fraction of the available RNAP core enzyme would be associated with σ^{70}, allowing for an increase in the amount of σ^{32}-containing RNAP holoenzyme in the cell and upregulation of σ^{32}-controlled genes. MglA may therefore exert some of its negative effects on gene expression by influencing the competition between the two available σ factors in the cell. Crl, a transcription factor in *E. coli,* promotes σ^{38} activity by

promoting formation of the σ^{38}-containing RNAP holoenzyme, and provides a precedent in the literature for a protein that promotes the formation of a specific holoenzyme species (Banta et al., 2013; Gaal et al., 2006; Pratt and Silhavy, 1998). Note that although it is formally possible that some of the effects of MglA on gene expression might be accounted for by the effects of MglA on cell growth, a mutant containing a transposon insertion in the FTL_0951 locus, which grows more poorly than a *mglA* mutant, did not exhibit increased expression of σ^{32}-regulated genes (Charity et al., 2007).

It is unclear to what extent MglA and SspA might influence the expression of σ^{70}-dependent genes by promoting the formation of the σ^{70}-containing RNAP holoenzyme. Regardless of whether or not the MglA-SspA complex specifically promotes formation of the σ^{70}-containing RNAP holoenzyme, the principal role of the MglA-SspA complex in *F. tularensis* with respect to virulence gene expression appears to be to provide a point of contact between PigR and RNAP. Consistent with this notion, PigR and MglA appear to be equally important for intracellular growth. Strains lacking either protein are equally defective for intramacrophage growth and for virulence in a mouse model of infection (Brotcke and Monack, 2008; Charity et al., 2009).

Our findings concerning the locations of PigR, MglA, and SspA together with our identification of a possible PRE have lead to a revised working model for how PigR works in concert with the MglA-SspA complex to positively regulate the expression of a specific set of genes. According to this model, PigR is associated with all σ^{70}-dependent promoters through its interaction with the RNAP-associated MglA-SspA complex. However, only at those promoters that contain a PRE does PigR make sufficiently strong contact with the DNA to further stabilize the binding of RNAP and activate transcription.

While our current working model for how PigR works in concert with the MglA-SspA complex (Figure 2.10) is consistent with our data thus far, much work remains to both test and clarify it. Our future work will include explicitly testing whether the PRE renders a promoter

responsive to PigR, determining whether PigR is a DNA-binding protein, and determining whether the MglA-SspA complex inhibits formation of the σ^{32}-containing RNAP holoenzyme.

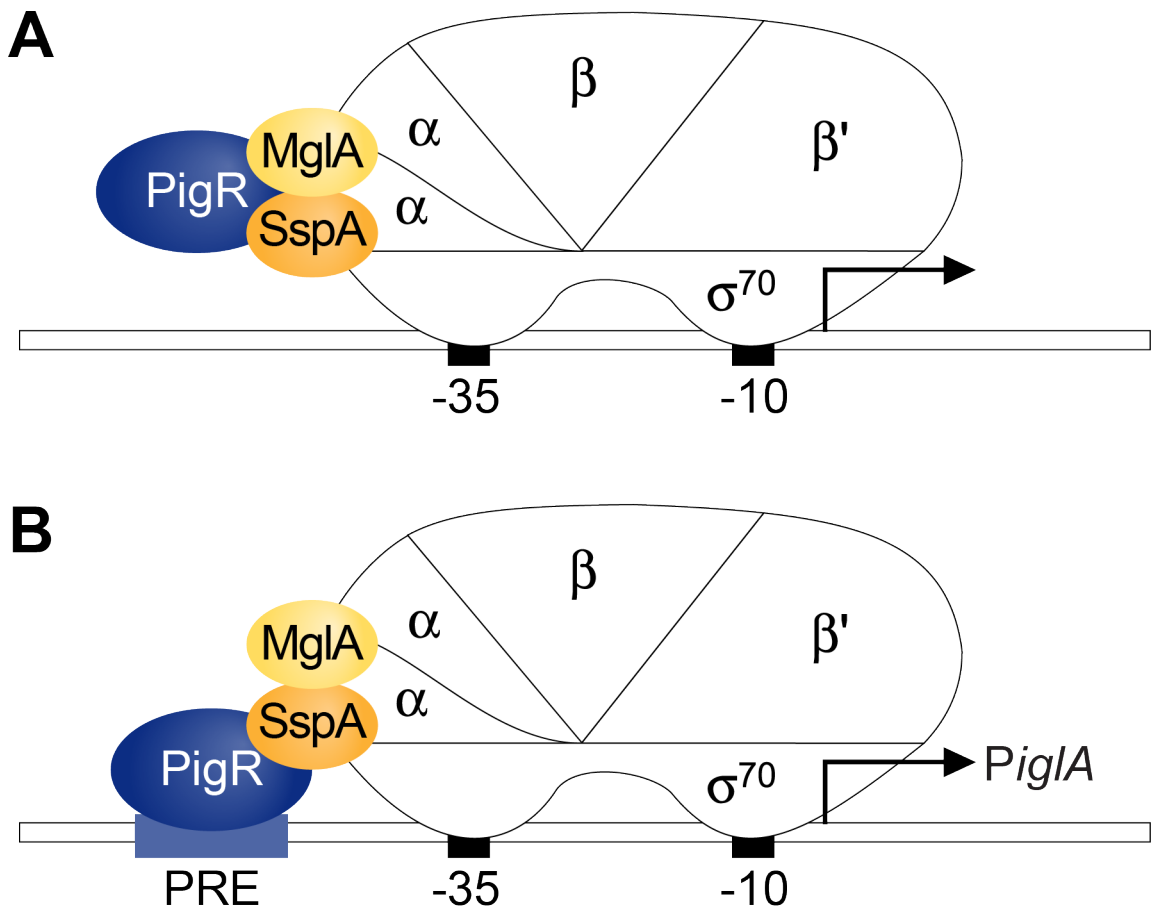

Figure 2.10. A revised working model for how the MglA-SspA complex and PigR control a common set of genes. A. At promoters not regulated by MglA, SspA, and PigR, the three factors are found at promoter regions through their interactions with each other and RNAP. **B.** At promoters regulated by MglA, SspA, and PigR such as the *iglA* promoter, PigR specifically recognizes DNA containing the PigR responsive element (PRE), interacts directly with the RNAP-associated MglA-SspA complex and stabilizes the binding of RNAP.

Materials and Methods

Plasmids, strains, and growth conditions

F. tularensis subsp. *holarctica* LVS was grown at 37°C in either Mueller Hinton (MH) broth (Difco), supplemented with glucose (0.1%), ferric pyrophosphate (0.025%), and Isovitalex (2%), or on cysteine heart agar (Difco) medium supplemented with 1% hemoglobin solution (VWR); when appropriate, kanamycin was used for selection at 5 µg/ml. *E. coli* strain XL1-blue (Stratagene) was used for plasmid construction and, when appropriate, kanamycin was used to select for resistance at 50 µg/ml.

Integration vectors

A modified version of pEX18Kan (provided by Shite Sebastian and Simon Dillon, Harvard Medical School, Boston, Massachusetts, United States) was used as the vector for integration constructs. We have used pEX18Kan for deletion constructs (Charity et al., 2007; 2009); it utilizes a ColE1 origin of replication, which is nonfunctional in LVS, and contains the T903 kanamycin resistance gene (Epicentre) driven by the LVS *groEL* promoter. The plasmid pKL01 was generated by first amplifying the last 400 base pairs (bp) of the FTL_1743 locus (*rpoC*), minus the stop codon, by PCR. The 5′ primer contained DNA specifying a KpnI site upstream of the gene fragment. The 3’ primer included DNA containing a NotI site and one extra base pair, encoding a 3 amino acid alanine linker. The linker is followed by DNA specifying the 11 amino acid vesicular stomatitis virus-glycoprotein (VSV-G) tag, followed by a stop codon and DNA specifying an EcoRI site. The corresponding PCR product was digested with KpnI and EcoRI and cloned into pEX18Kan that had been digested with KpnI and EcoRI, generating pKL01. We largely removed the *sacB* gene by digesting with MscI and EcoRV and re-ligating the vector together, resulting in pKL02.

Integration constructs for the remaining loci were generated by amplifying the last 250-400 bp (depending on gene size) of the gene using a 5′ primer containing a KpnI site and a 3′ primer containing a NotI site, which allows each fragment to be fused with DNA specifying the alanine linker and VSV-G epitope tag. Fragments were subcloned into pKL02 that had been digested with KpnI and NotI. An alternate integration plasmid was generated for genes whose putative operons would potentially be interrupted by plasmid integration. To do this, another *groES* promoter was amplified from LVS genomic DNA and cloned upstream of the gene fragment, into the BamHI and PstI sites of the vector derived from pKL02, to drive expression of downstream genes. This alternate integration construct was necessary in adding an epitope tag to the *sspA* gene. We did not successfully ChIP-Seq MglA-VSV-G, and used the LVS strain expressing MglA with a C-terminal TAP tag at native levels, described previously (Charity et al., 2007).

Strain construction

Electroporation of integration plasmids into LVS was performed as described in Maier et al., 2004. Cells in which a single homologous recombination event had occurred between the integration vector and the chromosome were selected on cysteine heart agar with 1% hemoglobin and 5 μg/ml kanamycin. Strains containing the correct integration were confirmed by colony PCR, by Western blotting and/or Southern blotting.

Southern blotting

Southern blots were largely performed as described in the Roche DIG protocols. Probes were synthesized using the PCR DIG Probe Synthesis Kit (Roche). For probes with AT-content

higher than 63%, amplified unlabeled probe was used as template for the probe labeling reaction and half the specified DIG dNTPs were used, supplemented with additional dATP and dTTP (for a final concentration of 100 μM dCTP and dGTP, 300 μM dATP, 265 μM dTTP, and 35 μM DIG-11-dUTP).

2.5 ug of genomic DNA was digested for 1 hour and separated on a 1% agrarose gel. The gel was incubated in 0.25 M HCl for 7 minutes, washed twice with deionized water, and incubated in Denaturation Solution (1.5 M NaCl, 0.5 M NaOH) twice for 20 minutes. DNA was transferred overnight to positively charged nylon membrane by capillary action and crosslinked to the membrane. Membranes were incubated in UltraHyb (Ambion) at the appropriate hybridization temperature for 1 hour prior to overnight hybridization at the same temperature in UltraHyb containing the appropriate probe. After hybridization, membranes were washed twice for 5 minutes in Low Stringency Buffer (2x SSC, 0.1% SDS) and twice for 15 minutes at 65°C in High Stringency Buffer (0.5x SSC, 0.1% SDS). The DIG Wash and Block Buffer Set (Roche) was used according to instructions to detect the DIG-labeled probe using CDP-Star (Roche).

Ectopic PigR expression

Plasmids pF and pF-PigR-V (described previously, Charity et al., 2007) were used as a negative control vector and to drive ectopic expression of PigR with a C-terminal VSV-G tag, respectively. The pF-PigR-V plasmid contains DNA encoding the PigR protein fused to the VSV-G epitope, which is driven from the *groES* promoter; the pF plasmid does not contain the *pigR* gene or DNA encoding the VSV-G epitope tag. These plasmids were electroporated into either the previously described LVS Δ*pigR* mutant strain (Charity et al., 2009), or a LVS Δ*pigR* Δ*mglA* mutant strain, which was created by using the pEX2-Δ*mglA* vector (Charity et al., 2007) in the Δ*pigR* background, by allelic exchange and confirmed by Southern blotting.

Chromatin Immunoprecipitation (ChIP)

Cells were grown at 37°C in 100 mL of supplemented MH to mid-log (OD_{600} 0.3-0.4), and when indicated, rifampicin (Sigma) was added to a final concentration of 50 µg/mL for 30 minutes before crosslinking. Cells were incubated in a final concentration of 1% formaldeyhyde (Sigma) for 30 minutes to crosslink, after which glycine (Sigma) was added to a final concentration of 250 mM. ChIP was performed in biological triplicate (excepting β′ + rifampicin, OxyR, and Δ*pigR* pF-PigR-V, which were performed in duplicate, and σ^{70} which was performed in quadruplicate) with either 40 mL or 80 mL of culture using anti-VSV-G agarose beads (Sigma, catalog no. A1970) for cells synthesizing VSV-G tagged transcription factors or IgG Sepharose beads (GE Healthcare, catalog no. 17-0969-01) for cells synthesizing TAP-tagged transcription factors essentially as described previously (Castang et al., 2008), except that a water bath sonicator (Biorupter, Diagenode) was used to lyse cells and shear chromosomal DNA to 200 to 500 bp. Immunoprecipitated DNA was quantified using the Quant-iT™ PicoGreen® dsDNA Assay Kit (Invitrogen). The same protocol was performed with the untagged LVS strain as a mock immunoprecipitation (mock IP) control.

Illumina Library Preparation

Libraries were constructed with approximately 2 to 160 ng immunoprecipitated DNA using the TruSeq DNA Sample Prep Kit (Illumina, catalog no. FC-121-2002), generally following the supplied protocol; adapters were diluted 1:10 before ligation and libraries were gel-purified after 11 cycles of amplification. Libraries were quantified using the Quant-iT™ PicoGreen® dsDNA Assay Kit (Life Technologies) and sequenced by Elim Biopharmaceuticals, Inc. (Hayward, CA), using an Illumina Genome Analyzer IIx generating 36 bp single-end reads.

ChIP-Seq data analysis

For each strain, the 36 bp reads were mapped to the *F. tularensis* subsp. *holarctica* LVS genome (NCBI locus AM233362) and the sequence of the integrated plasmid, if applicable, using bowtie2-2.0.6 (Langmead and Salzberg, 2012), allowing up to one mismatch. Regions of enrichment were called using QuEST, version 2.42 (Valouev et al., 2008). The three mock IP biological replicates, consisting of approximately 30.6 million reads, were merged and used as a background control for each biological replicate. Peaks in each biological replicate are regions that fit the following criteria: they are 1.5-fold enriched for reads over background, with a positive peak shift and strand correlation, and a q-value of less than 0.01. Peaks for each immunoprecipitated protein were defined as the maximal region identified in at least two biological replicates. Tracks were visualized using the Integrative Genomics Viewer (IGV), version 2.3 (Thorvaldsdóttir et al., 2013). Peak analyses were carried out using Perl scripts and BEDtools (Quinlan and Hall, 2010).

Immunoblots

Cell lysates were separated by SDS-PAGE on 4-12% or 12% Bis-Tris NuPAGE gels in MES or MOPS running buffer (Life Technologies). Either the iBlot dry blotting system or the XCell II Blot Module (Life Technologies) was used to transfer proteins to either PVDF or nitrocellulose. Membranes were blocked with SuperBlock Blocking Buffer (Pierce) with 0.25% Surfact-Amps 20 (Pierce) for 1 hour to overnight. Membranes were then probed with polyclonal anti-VSV-G (Sigma) or anti-GroEL (provided by Karsten Hazlett, Albany Medical College, Albany, New York, United States) for one hour, washed (10 minutes incubations in TBST plus 0.25% Surfact-Amps 20, 4 times) and re-blocked for 1 hour. After membranes were incubated with polyclonal goat anti-rabbit and washed, proteins were detected using SuperSignal West Pico Chemiluminescent Substrate or SuperSignal West Femto Chemiluminescent Substrate (Pierce).

Transcriptomic Analysis

Cells of LVS, LVS Δ*mglA* (described in Charity et al., 2007), and LVS Δ*pigR* (described in Charity et al., 2009) were grown to mid-log in biological triplicate. 1 mL of each sample was pelleted (20,000 rcf for 5 minutes), resuspended in 500 µL Qiagen buffer RLT, frozen on dry ice and stored at -80°C. Equal amounts of lysate, normalized to OD_{600}, in 4 µL Qiagen buffer RLT and 1 µL water were submitted to the Epithelial Cell Biology Core Facility (Boston Children's Hospital) for processing using the Nanostring nCounter Prep Station and Digital Analyzer according to the manufacture's instructions. Total transcript counts were normalized using internal controls with background subtraction. Transcript abundance was determined by averaging biological triplicates. Criteria indicating significant change in gene expression are a 2-fold change in transcript abundance and $P < 0.05$ in a two-tailed Student's t-test.

Identifying a potential PRE by MEME

Genes with significant changes in expression in LVS Δ*pigR* in comparison to LVS from either this work (Table 2.2) or Charity et al., 2009 were examined for promoter regions with detectable PigR, identifying 11 genes (FTL_0026, FTL_0111, FTL_0126, FTL_0131, FTL_0207, FTL_0449, FTL_0491, FTL_1218, FTL_1219, FTL_1678, FTL_1790). The 400 bp region surrounding the maximal PigR binding site, upstream from 11 genes, excluding coding regions, was searched for a common motif using MEME (Bailey and Elkan, 1994).

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Chapter 3

Architecture of the virulence control network in *Francisella tularensis*

Author contributions: All of the work in this chapter was performed by Kathryn Levasseur, with the following exceptions. Cathy Su generated 10 plasmids for epitope-tagging transcription factors, 2 *F. tularensis* LVS strains that produce an epitope-tagged transcription factor and performed 2 ChIP-Seq experiments.

Abstract

Studies of *Francisella tularensis* virulence regulation have focused on a few transcription regulators critical for intracellular survival. Yet many other transcription regulators have been identified as virulence factors and very little is known about how they function to promote pathogenesis. In this study, we identify the locations on the genome at which thirty *F. tularensis* transcription factors are found, including all of those previously implicated in the control of virulence. For each factor, we have described a putative regulon, and, in the case of OxyR and the LysR-type transcription regulator FTL_0040, we have determined which regulon members have previously been implicated in virulence. We did not detect PmrA, a transcription factor demonstrated to promote expression of genes on the *Francisella* Pathogenicity Island (FPI), at critical FPI promoters. We suggest that PmrA may be indirectly influencing the expression of FPI genes by directly controlling the expression of another factor known to affect FPI gene expression, PigR. While genes on the FPI are upregulated under low iron conditions, a characteristic typical of Fur-regulated genes, we did not detect Fur at any promoter on the FPI. Based on their locations, we suggest that three proteins, PmrA, IclR, and the LysR-type transcription regulator FTL_1568, have a non-cannonical function, potentially as nucleoid-associated proteins. Finally, we have constructed a model of the network of transcription factors involved in controlling virulence. This network model reveals a potential hierarchy of regulation in which FTL_0040 is implicated in the control of a large subset of transcription factors necessary for virulence.

Introduction

Francisella tularensis is a Gram-negative, intracellular bacterium and is the etiological agent of tularemia. While it mostly causes disease in small mammals, humans can be infected via a number of routes. *F. tularensis* is extremely infectious and inhalation of less than ten aerosolized bacteria can cause disease (Tärnvik and Chu, 2007). In humans, two subspecies of *F. tularensis* cause disease; infection with *F. tularensis* subsp. *holarctica* results in relatively mild disease while infection with *F. tularensis* subsp. *tularensis* can cause a disease with much greater severity (Ellis et al., 2002). An attenuated derivative of the *F. tularensis* subsp. *holarctica* strain, the Live Vaccine Strain or LVS, is unable to cause disease in humans but maintains virulence in animal models (Eigelsbach and Downs, 1961), and we have used this strain in our work (which we will subsequently refer to simply as LVS). Another subspecies, *F. tularensis* subsp. *novicida* (referred to here as *F. novicida*), is also commonly used in research due to its inherent inability to infect immunocompetent individuals and its ability to cause disease in animal models. However, the combination of the highly infectious nature of *F. tularensis*, the multiple routes of infection, and the potential of some strains to cause lethal disease has lead the United States government to consider *F. tularensis* a potential bioweapon (Gallagher-Smith et al., 2004). Despite much recent study, there are still significant gaps in our knowledge of the molecular mechanisms underlying the infectious and virulent nature of *F. tularensis*.

As an intracellular pathogen, the ability to infect a host cell and subsequently survive and replicate is essential for *F. tularensis* virulence. It is clear that a particular set of genes, thought to encode a type VI-like secretion system, is required for intracellular survival and growth. This group of genes is found at the genetic locus termed the *Francisella* Pathogenicity Island (FPI). The FPI is found in all the *Francisella* species sequenced thus far and it appears to have been horizontally acquired as its GC-content is significantly lower than the rest of the genome (Bröms et al., 2010; Nano et al., 2004).

Several transcription factors have been implicated in the control of genes present on the FPI. MglA and SspA are *F. tularensis* stringent starvation protein A (SspA) family members which form a heteromeric complex that interacts directly with RNA polymerase (RNAP) to promote the expression of virulence genes, including all those found on the FPI (Baron and Nano, 1998; Charity et al., 2007). Another protein, termed PigR in LVS (FevR in *F. novicida*), interacts with the MglA-SspA complex to coordinate virulence gene expression (Charity et al., 2009). A small molecule, ppGpp, is also found to control essentially the same set of genes as PigR, MglA, and SspA (Charity et al., 2009). Working in concert with PigR and the MglA-SspA complex, ppGpp is thought to promote the interaction between the MglA-SspA complex and PigR (Charity et al., 2009). PmrA is an orphan response regulator in *F. tularensis* that has been demonstrated to regulate a subset of the MglA/SspA/PigR regulated genes and is proposed to function through a direct interaction with MglA and SspA at FPI promoters (Bell et al., 2010; Mohapatra et al., 2007; Dai et al., 2010).

While it is clear that control of the FPI and regulation by MglA, SspA, PigR, and PmrA is critical for virulence, many additional predicted transcription factors have been shown to be important for virulence. These additional transcription factors encompass most of the transcription factors present in the *F. tularensis* genome (Table 1), and in most cases it is unclear how they control virulence. Despite widespread identification of transcription factors other than MglA, SspA, PigR, and PmrA as virulence determinants, few studies have attempted to address how these less well-studied transcription factors affect virulence. It is not uncommon to see virulence regulators coordinated in a network or hierarchy to control virulence factor expression; there is no such network or hierarchy described in *F. tularensis*. Yet the information regarding how genes critical for virulence are coordinately regulated can provide further insight into the molecular mechanisms underlying pathogenesis.

Using chromatin immunoprecipitation followed by high-throughput sequencing (ChIP-Seq) (Robertson et al., 2007), we determined the direct targets of 30 transcription factors,

including all of those previously implicated in virulence in *F. tularensis*. Our findings suggest that in LVS, PmrA controls the expression of FPI genes indirectly and that PmrA, IclR and FTL_1568 may function as nucleoid-associated proteins (NAPs). Furthermore, we have connected specific virulence regulators to the control of specific virulence genes, providing an explanation for the roles of several regulators in the control of virulence. Finally, we present a model for the regulatory network controlling virulence gene expression in *F. tularensis* on a genome-wide scale.

Results

Selection of putative transcription factors for ChIP-Seq analysis

We sought to determine the binding sites, on a genome-wide scale, of all the transcription factors in *F. tularensis* that have been implicated in controlling virulence (Table 3.1). To do this we performed ChIP-seq of each putative virulence regulator (Park, 2009; Robertson et al., 2007). In particular, we created strains of LVS that synthesized each of the 30 transcription factors with a vesicular stomatitis virus-glycoprotein (VSV-G) epitope tag or the tandem affinity purification (TAP) epitope tag fused to its C-terminus, according to the scheme outlined in Figure 3.1. Because these strains were created by modifying the native copy of each gene specifying a particular transcription factor, each factor we analyzed by ChIP-Seq was produced at close to wild-type levels. ChIP-Seq was performed with cells grown to mid-log in supplemented Mueller-Hinton broth, conditions under which FPI-encoded genes that are important for virulence are expressed (Charity et al., 2007).

Table 3.1. Transcription factors analyzed by ChIP-Seq

Locus	Gene Name	Gene Product	Essential[1]	Report of Virulence-Related Phenotype
FTL_1743	*rpoC*	DNA-directed RNA polymerase, β′ subunit	+	-
FTL_1050	*rpoD*	RNA polymerase σ^{70} factor	+	-
FTL_0851	*rpoH*	RNA polymerase σ^{32} factor	+	(Grall et al., 2009)
FTL_0616	*rpoA2*	DNA-directed RNA polymerase subunit α2	-	(Asare and Abu Kwaik, 2010; Asare et al., 2010)
FTL_1474	*greA*	Transcriptional elongation factor	-	(Asare and Abu Kwaik, 2010; Asare et al., 2010; Su et al., 2007)
FTL_1185	*mglA*	Macrophage growth locus, subunit A	-	(Baron and Nano, 1998; Moule et al., 2010)
FTL_1606	*sspA*	Stringent starvation protein A	-	(Asare and Abu Kwaik, 2010; Asare et al., 2010; Charity et al., 2007)
FTL_0449	*pigR*	Transcription factor PigR	-	(Brotcke and Monack, 2008; Charity et al., 2009; Wehrly et al., 2009)
FTL_0428	*parB*	Chromosome partition protein B	-	(Su et al., 2007)
FTL_0552	*pmrA*	Two-component response regulator	-	(Asare and Abu Kwaik, 2010; Asare et al., 2010; Bell et al., 2010; Mohapatra et al., 2007; Moule et al., 2010)
FTL_1364	*iclR*	IclR family transcriptional regulator	-	(Asare and Abu Kwaik, 2010; Kraemer et al., 2009; Weiss et al., 2007)
FTL_1014	*oxyR*	Oxidative stress transcriptional regulator	-	(Moule et al., 2010)
FTL_1831	*fur*	Ferric uptake regulation protein	-	(Deng et al., 2006)
FTL_0662	*lexA*	Prophage repressor protein	-	(Asare et al., 2010)
FTL_1276	*birA*	Bifunctional protein BirA	-	(Napier et al., 2012)
FTL_1126	*hipB*	Transcriptional regulator	-	(Asare et al., 2010)
FTL_0370	*arsR*	Arsenical resistance operon repressor	-	(Asare et al., 2010)
FTL_1079	-	Helix-turn-helix family protein	-	(Moule et al., 2010)
FTL_1216	-	Hypothetical protein FTL_1216	-	(Asare et al., 2010)
FTL_1222	-	MarR family transcriptional regulator	-	(Asare et al., 2010)
FTL_1231	-	Transcriptional regulator	-	(Asare et al., 2010)
FTL_1293	-	Hypothetical protein FTL_1293	-	(Kraemer et al., 2009)
FTL_0040	-	LysR family transcriptional regulator	-	(Kraemer et al., 2009)
FTL_0062	-	LysR family transcriptional regulator	-	(Moule et al., 2010)
FTL_0742	-	LysR family transcriptional regulator	-	(Moule et al., 2010)
FTL_0844	-	LysR family transcriptional regulator	-	(Moule et al., 2010)
FTL_1176	-	LysR family transcriptional regulator	-	(Moule et al., 2010)
FTL_1193	-	LysR family transcriptional regulator	-	(Asare and Abu Kwaik, 2010; Asare et al., 2010; Moule et al., 2010)

Table 3.1. Transcription factors analyzed by ChIP-Seq, continued

FTL_1568	-	LysR family transcriptional regulator	-	-
FTL_1634	-	LysR family transcriptional regulator	-	-

[1]Based on data from *F. novicida*. "+", gene is reported as a potentially essential gene; "-", gene has not been reported as potentially essential (Gallagher et al., 2007).

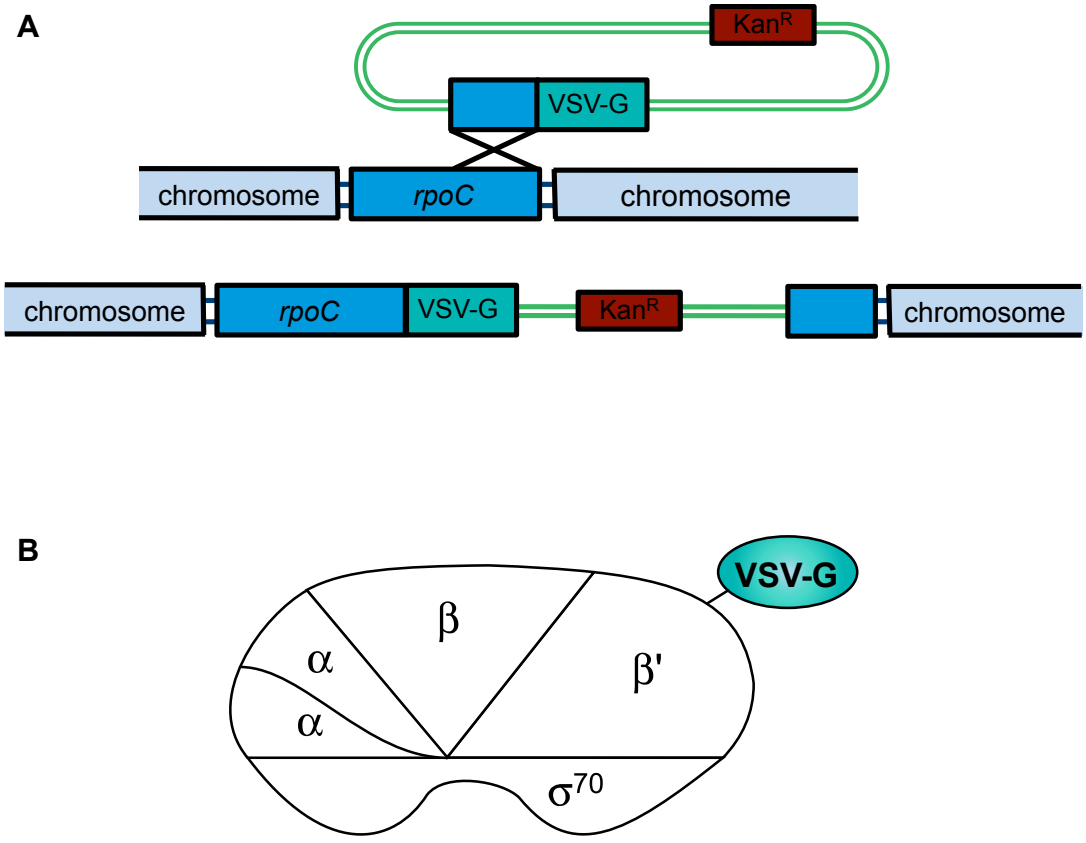


Figure 3.1. Schematic diagram for producing strains with epitope-tagged transcription factors. A. The integration vector contains a fragment of the *rpoC* gene (FTL_1743) followed by DNA specifying a VSV-G epitope tag. A single homologous recombination event produces a strain, LVS β′-V, that synthesizes the VSV-G tagged β′ subunit of RNAP (β′-V) from the native *rpoC* locus. **B.** The VSV-G tagged β′ subunit is incorporated into the RNAP holoenzyme complex.

Because the list of virulence-associated regulators includes 7 of the 9 total LysR family members, we also included the 2 LysR family members not previously implicated in virulence to our analyses in order to study the entire LysR family. Furthermore, in order to be able to relate the location of any transcription regulator to a particular promoter, we determined the locations of RNA polymerase (RNAP) in *F. tularensis*. Promoter regions are defined here as regions enriched upon ChIP-Seq of the β′ subunit of RNAP after treatment of cells with rifampicin (rif); rif effectively leads to trapping of the RNAP-holoenzyme complex at promoter regions. As a complementary approach to determining the locations of promoters we also determined the locations of the two σ factors present in *F. tularensis* (σ^{70} and σ^{32}) by ChIP-Seq (Campbell et al., 2001; Herring et al., 2005). After merging the overlapping peaks identified by each promoter-associated transcription factor, we found 582 putative promoter regions throughout the genome (see Appendix, Table S2). The majority of promoters identified are found for the most part in non-coding, intergenic regions (495 promoters, 85% of total). The AT-content of promoter regions is enriched relative to that of the rest of the genome; while AT-content across the entire genome is 67.8%, promoter regions have an AT-content of 71.8%.

We performed ChIP-Seq on 30 of the 44 putative transcription factors in LVS (Table 3.1 and Appendix, Table S1). The chart in Figure 3.2 illustrates, for each transcription factor, the number of ChIP-Seq peaks that overlap with promoter regions. Because we defined promoter regions as those enriched for the β′ subunit of RNAP after treatment of cells with rif, or either σ factor, by definition, all the ChIP-Seq peaks for those factors are promoter-associated.

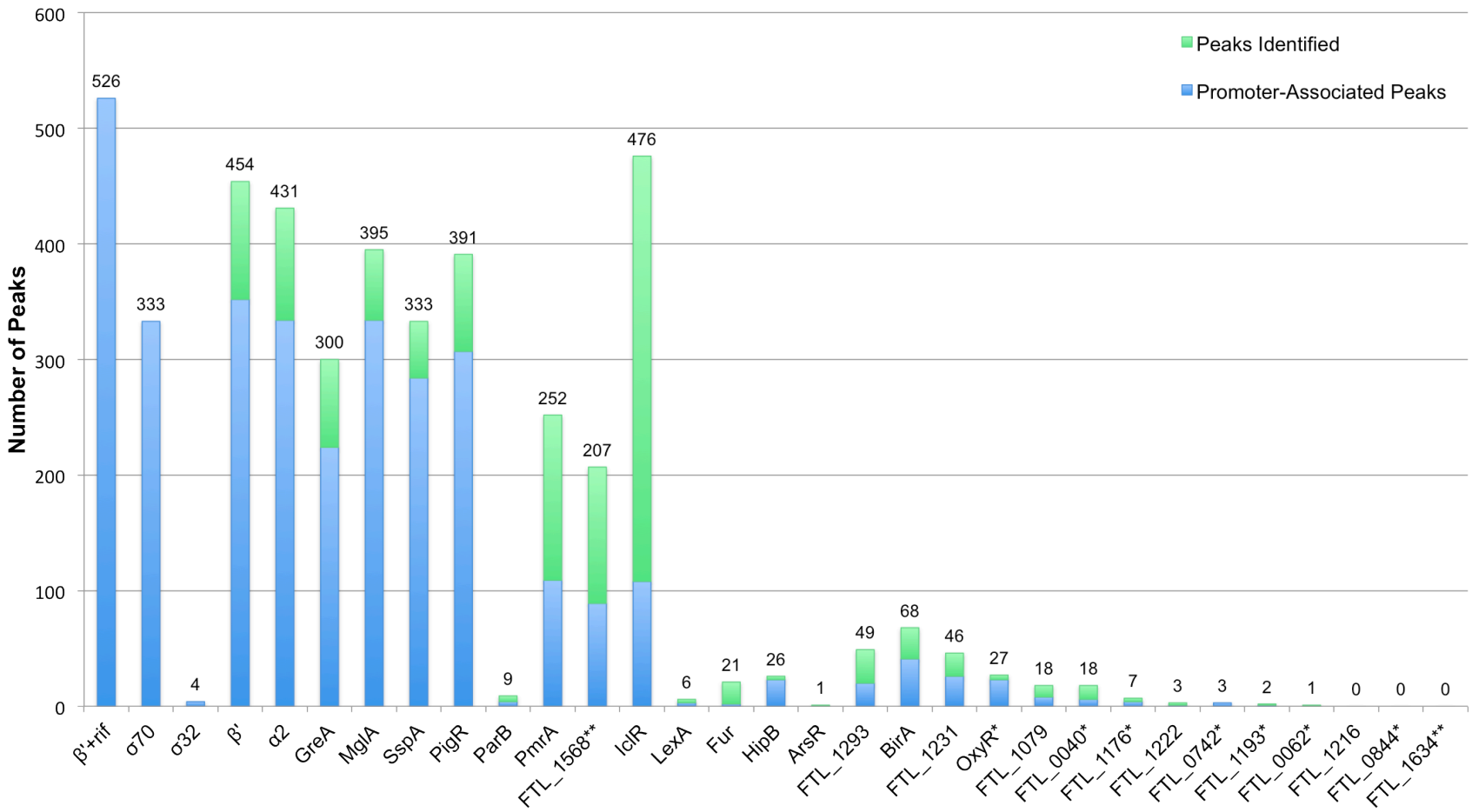


Figure 3.2. Number of peaks identified from ChIP-Seq of each *F. tularensis* transcription factor examined. Transcription factors are listed along the X-axis and the Y-axis represents the number of regions enriched for a particular transcription factor (peaks). Dark blue bars indicate peaks that overlap with promoter regions; green bars represent peaks that do not overlap promoter regions.

*LysR-type transcription regulators

**LysR-type transcription regulators not previously implicated in virulence

Figure 3.2 also illustrates that the large number of MglA, SspA, and PigR peaks and extent of their overlap with promoter regions reflects that these proteins are found at locations very similar to the locations at which RNAP is found (as evidenced by both the β′ subunit and α2 subunit) as well as locations at which the elongation factor GreA is found. This highlights that we identified MglA, SspA, and PigR at the majority of promoters, as described in Chapter 2. Our results from Chapter 2 suggest that the MglA-SspA complex may be present at all σ^{70}-dependent promoters, functioning to promote formation of the σ^{70}-containing RNAP holoenzyme and providing a point of contact for PigR, which promotes gene expression from specific promoters.

PmrA may indirectly control the expression of genes on the FPI and be a nucleoid-associated protein (NAP)

PmrA is a key regulator of virulence gene expression in *F. tularensis* and, in LVS, is the only known response regulator. PmrA is required for virulence in mice and controls the expression of many genes, exerting both positive and negative effects (Mohapatra et al., 2007; Sammons-Jackson et al., 2008). A subset of PmrA-controlled genes are also controlled by MglA, SspA, and PigR in both LVS and in *F. novicida*, including several of those on the FPI (Mohapatra et al., 2007; Sammons-Jackson et al., 2008). One model specifies that PmrA exerts its stimulatory effects on FPI-encoded genes directly—once bound to target promoters on the FPI, PmrA interacts with the RNAP-associated MglA-SspA complex to activate transcription (Dai et al 2011). Consistent with this model, purified PmrA has been shown to bind directly to the *pdpD* promoter from the *F. novicida* FPI, and MglA and SspA were found to co-precipitate with PmrA (Bell et al., 2010). However, in LVS, where there are two copies of the FPI, PmrA controls the expression of FPI genes even though the FPIs do not contain the *pdpD* promoter region previously shown to bind PmrA *in vitro*. Whether PmrA exerts its effects on expression of FPI-encoded genes directly in LVS was therefore not known.

To determine whether PmrA might influence expression of the FPI genes directly, we used ChIP-Seq to determine the location of PmrA. We identified 252 regions enriched for PmrA, ranging from 1.5 to 536-fold enriched. Within the FPI, we found that PmrA was associated with 3 regions, one immediately upstream of *pdpE*, one in the middle of *pdpC*, and the other upstream of *pdpB* (Appendix, Figure S1). We did not detect any association of PmrA with the *iglA* promoter (Figure 3.3) suggesting that in LVS PmrA does not exert its effects on expression of the *igl* genes by binding directly to the *iglA* promoter.

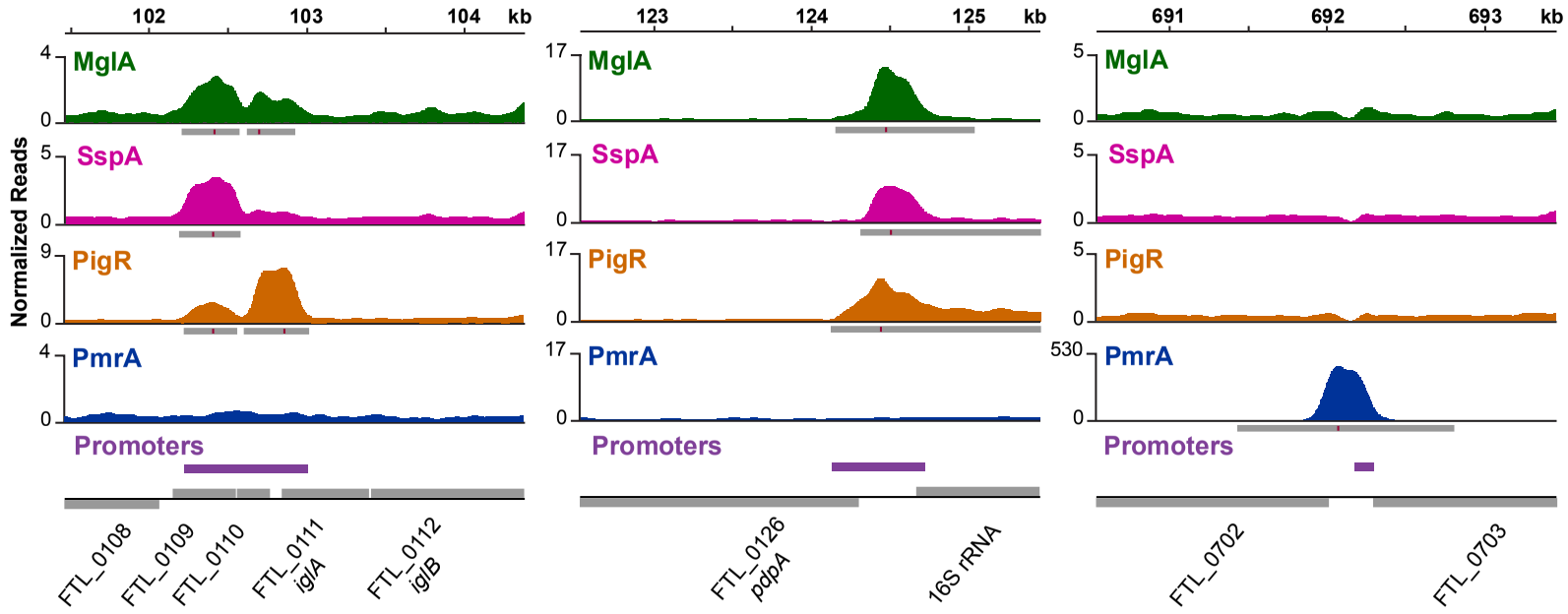


Figure 3.3. PmrA is not detected at the *iglA* or *pdpA* promoters but is highly enriched at the FTL_0702 promoter. Genomic position is indicated on the X-axis by *F. tularensis* LVS locus number; gray boxes represent genes, which are above the black line if they are encoded on the plus strand and below the black line if they are encoded on the minus strand. Representative tracks illustrating the density of normalized mapped sequencing reads after ChIP-Seq of MglA (green), SspA (pink), PigR (orange), and PmrA (blue) are depicted on the Y-axis. Areas of significantly enriched reads, referred to here as peaks, are indicated by the horizontal gray boxes below the read density plot and sites of maximum enrichment are indicated by the red lines. Putative promoter regions, which are areas with significant enrichment of β′ + rif, σ^{32}, or σ^{70} association with the chromosome, are indicated by the purple boxes above the gene annotations. There is no detectable enrichment of PmrA at either the *iglA* (left panel) or *pdpA* (center panel) promoters, but PmrA is detected at the FTL_0702 promoter region (right panel).

PmrA has been shown to positively regulate expression of the *pigR* gene (Mohapatra et al., 2007; Sammons-Jackson et al., 2008). The FPI genes are also positively regulated by PigR, MglA and SspA, all three of which are found at the promoters of the FPI genes by ChIP-Seq (Figure 3.3). The results depicted in Figure 3.4 show that PmrA is detected at the *pigR* promoter and appears to have another site of significant enrichment at the 3' end of the *pigR* gene. While the significance of PmrA enrichment downstream from the *pigR* promoter region is unclear, the presence of PmrA at the *pigR* promoter suggests that PmrA may be directly regulating *pigR* gene expression. Taken together, our findings suggest that in LVS, PmrA does not regulate expression of the *igl* genes on the FPIs directly. Rather, PmrA likely positively influences the expression of the *igl* genes, together with other members of the MglA/SspA/PigR regulon, by directly influencing the expression of *pigR*.

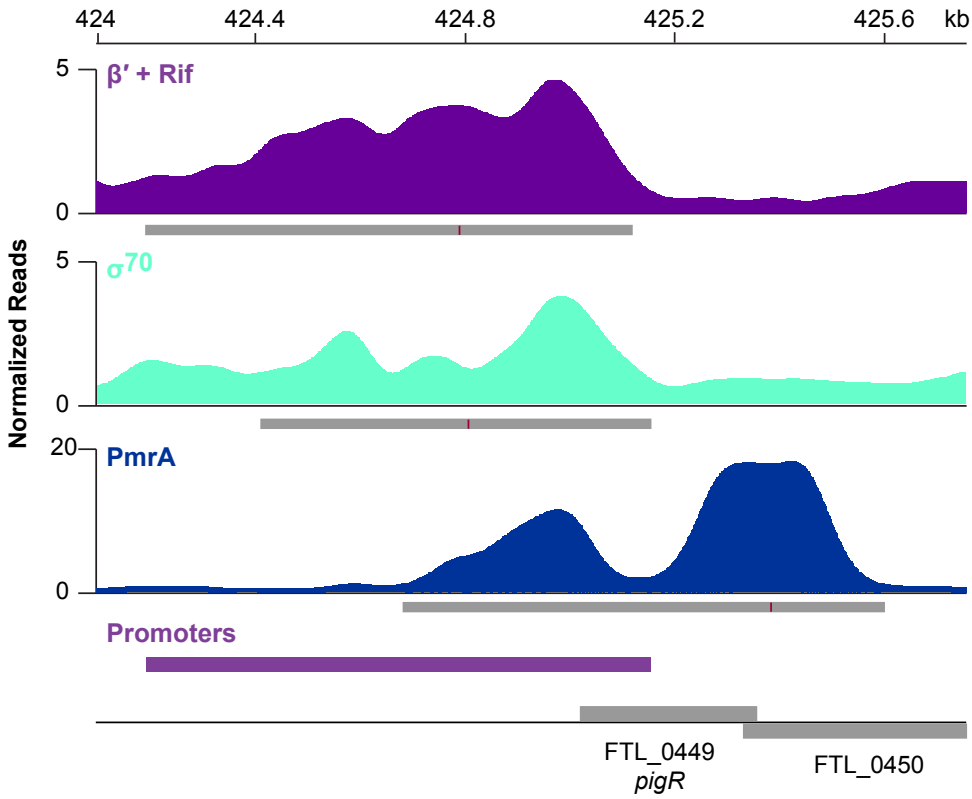


Figure 3.4. PmrA is found at the *pigR* promoter. Genomic position is indicated on the X-axis by *F. tularensis* LVS locus number; gray boxes represent genes, which are above the black line if they are encoded on the plus strand and below the black line if they are encoded on the minus strand. A representative track illustrating the density of normalized mapped sequencing reads after ChIP-Seq of each factor is depicted on the Y-axis. Areas of significantly enriched reads, referred to here as peaks, are indicated by the horizontal gray boxes below the read density plot and sites of maximum enrichment are indicated by the red lines. Putative promoter regions, which are areas with significant enrichment of β′ + rif, σ^{32}, or σ^{70} association with the chromosome, are indicated by the purple horizontal boxes above the genomic position markers. PmrA (dark blue track) is significantly enriched at the *pigR* promoter.

The region with the highest enrichment of PmrA is found close to the FTL_0702 gene, whose expression is known to be negatively regulated by PmrA (Mohapatra et al., 2007; Sammons-Jackson et al., 2008). It therefore appears likely that PmrA functions directly as a repressor of the FTL_0702 gene.

Of the regions we identified as associated with PmrA, only 21% were found to intersect regions enriched for both MglA and SspA. Conversely, 74% of the regions associated with PmrA were not enriched for either MglA or SspA. This is in sharp contrast to our findings with PigR, a protein which has been demonstrated to interact directly with the MglA-SspA complex (Charity et al., 2009); 69% of the regions identified as enriched for PigR are also enriched for both MglA and SspA (see Chapter 2). The finding that the MglA-SspA complex is not found at many of the same locations as PmrA suggests that PmrA does not function at all locations through direct contact with the MglA-SspA complex.

PmrA may not function as a canonical transcription factor at every location. Unlike MglA, SspA, or PigR, PmrA is found associated with many non-promoter regions; fewer than half of the peaks enriched for PmrA, 43%, are found at promoter regions (Figure 3.5). The lack of overlap between PmrA enrichment and promoters is significantly different from the approximately 80% or more of the regions identified by ChIP-Seq of MglA, SspA, or PigR associated with promoters (Figure 3.5). Because our method of defining a promoter region relies on our ability to detect the presence of RNAP, it is possible that the presence of PmrA at the 143 non promoter-associated locations is indicative of PmrA functioning as a repressor at these locations by occluding RNAP. However, of the PmrA-associated regions not associated with promoters, approximately 47% are found entirely within coding regions, a non-canonical location for promoter sequences. Additionally, genes associated with regions of PmrA enrichment (genes with translational start sites within 1 kb of the peak maximum) do not appear to have similar functions, as determined by cluster of orthologous group (COG) categories or by KEGG pathway analysis. Indeed, the abundance and diversity of PmrA ChIP-Seq peaks are

less suggestive of PmrA functioning solely as a canonical transcription factor and raise the possibility that PmrA may be more akin to a nucleoid-associated protein (NAP, Dillon and Dorman, 2010) with the potential to play a role in chromosome organization.

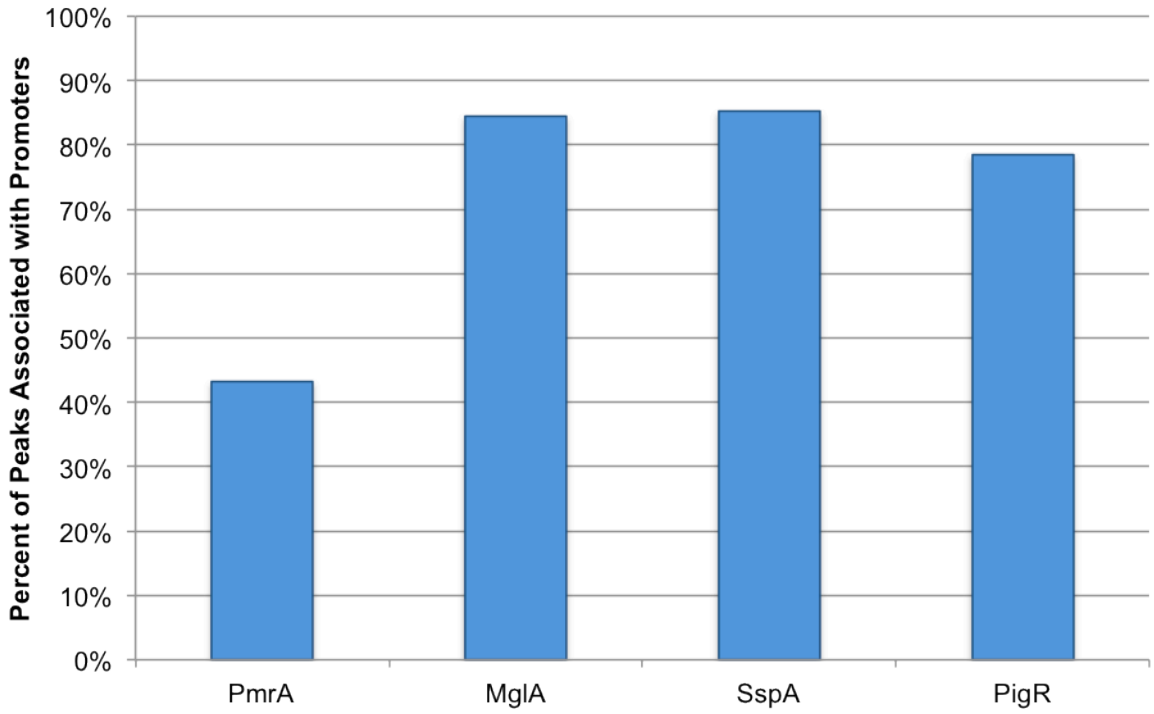


Figure 3.5. PmrA ChIP-Seq peaks are less frequently associated with promoters than are ChIP-Seq peaks of MglA, SspA, or PigR. The Y-axis indicates the percent of regions identified as enriched that intersect with promoters. Specific factors are labeled on the X-axis. Less than 50% of the peaks identified by ChIP-Seq of PmrA are promoter-associated, while approximately 80% or more of the peaks identified by ChIP-Seq of MglA, SspA, and PigR are promoter-associated.

Identification of three potential NAPs in *F. tularensis*

Our ChIP-Seq analysis of PmrA suggest that PmrA may exert its effects as a transcription factor, at least in some instances, in a non-canonical manner, potentially through influencing the organization of the chromosome as a NAP. We suggest that PmrA may function in part as a NAP based on several observations: PmrA-associated regions are numerous, are found at both intergenic and intragenic locations (Figure 3.6A), and lastly, PmrA-enriched peaks are not more likely to be found at promoter regions than non-promoter regions; that is to say, it appears equally likely that highly enriched peaks would be found at non-promoter locations as at promoter locations (Figure 3.6B).

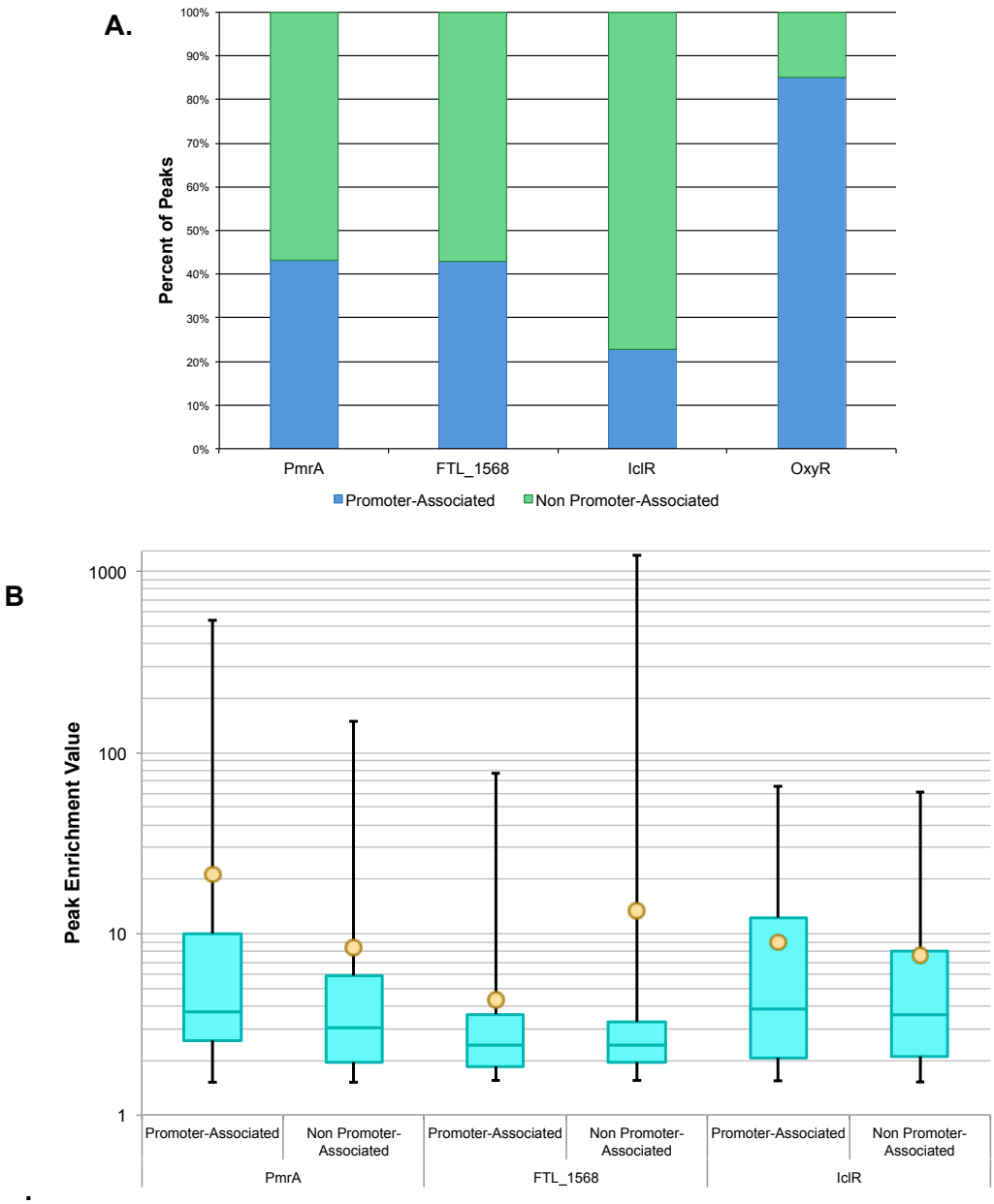


Figure 3.6. PmrA, FTL_1568, and IclR are found at promoter and non-promoter regions. A. Percent of regions identified as enriched that intersect with promoters. Specific factors are labeled on the X-axis. Less than 50% of the ChIP-Seq peaks identified by ChIP-Seq of PmrA, FTL_1568, and IclR are promoter-associated, while more than 80% of OxyR ChIP-Seq peaks are associated with promoters. **B.** Enrichment at peaks associated with promoters is not greater than at peaks not associated with promoters. For each category, box and whisker plot depicts the maximum enrichment (upper range), minimum enrichment (lower range), the median (center dark blue line), first (bottom of blue box), and third quartile (top of blue box) values, as well as the average (round yellow marker). There is no significant difference in enrichment between the promoter-associated and non promoter-associated peaks as assessed by Mann-Whitney U-test ($p > 0.01$).

In addition to PmrA, we found two other predicted transcription factors that have ChIP-Seq peaks with similar characteristics: IclR and the LysR-type transcription regulator FTL_1568. We found that the LysR-type transcription regulator FTL_1568 is associated with 207 genomic locations, with regions of association ranging in enrichment from 1,223 to 1.5-fold. Of these, 89 (43%) intersect with promoter regions (Figure 3.6A). Thus, the majority of FTL_1568 ChIP-Seq peaks appear to not be promoter-associated. It should be noted that it is possible that what we detect as a non-promoter associated ChIP-Seq peak is actually at a promoter region, but that under the conditions examined, the promoter is not active; in fact, a promoter region may specifically be inactive due to the binding of a transcription factor occluding RNAP. An inactive promoter is more likely to be the case for either ChIP-Seq peaks found at intergenic regions that have not been identified as promoters, or at regions found entirely within coding regions and directly upstream from an annotated translational start site. Most of the non-promoter associated ChIP-Seq peaks identified for FTL_1568, 64 of the 118 (54%), are found entirely within coding regions, less suggestive of association with a promoter and more suggestive of an alternate function for FTL_1568.

In comparison to PmrA and FTL_1568, the genomic regions of IclR association are similarly diverse and even more abundant. We identified 476 IclR ChIP-Seq peaks, which is second in terms of abundance only to promoter regions identified by ChIP-Seq of the β′ subunit of RNAP in the presence of rif. Regions associated with IclR ranged from 1.5 to 66-fold enriched for the factor. Approximately 23% (109) of these IclR-associated regions are found to intersect with promoter regions (Figure 3.6A). The IclR ChIP-Seq peaks that are not associated with promoter regions are mostly (262 of 368 or 71%) not found in intergenic regions, that is, they are located entirely within coding regions. Additionally, the AT-content of IclR peaks appears to reflect the overall AT-content of the genome rather than of promoter regions, as peak regions are found to consist of 66.4% AT while promoter regions are higher, at 71.8% AT.

As is the case with PmrA, the promoter-associated peaks of FTL_1568 or IclR are not significantly more enriched for each respective factor than non-promoter-associated peaks (Figure 3.6B). Also like PmrA, there does not appear to be any significant enrichment of COG categories or KEGG pathways associated with genes that are found within 1kb of the point of most enrichment for ChIP-Seq peaks identified for either IclR or FTL_1568.

We have demonstrated that PmrA, FTL_1568, and IclR are all found associated with numerous and varied genomic locations and they do not appear to be significantly enriched in association with a particular class of genes or genes implicated in a particular function. Our findings may be consistent with PmrA, FTL_1568, and IclR not only functioning as classical transcription activators or repressors but also functioning more generally as factors used to organize the chromosome.

Fur may not control expression of FPI-encoded genes directly

Previous studies have suggested that Fur, the ferric uptake regulator, may control the expression of genes on the FPI directly (Deng et al., 2006; Lenco et al., 2007). Under iron-replete conditions, Fur binds its co-factor Fe^{2+}, allowing it to bind the DNA. Fur typically acts as a repressor, binding to promoter DNA and occluding RNAP. Under conditions of iron limitation, Fur is no longer associated with Fe^{2+} and dissociates from the DNA to allow transcription. Fur is a site-specific DNA-binding protein and the site on the DNA to which Fur binds is referred to as a Fur box, as it contains a specific sequence motif. A sequence with homology to the Fur box identified in *E. coli* was found upstream of the FPI-encoded *pdpB* and *iglC* genes (Deng et al., 2006) and when cells are grown in iron-restricted conditions, expression of the FPI genes is induced (Buchan et al., 2009; 2008; Deng et al., 2006; Lenco et al., 2007). This suggests that FPI gene expression may be controlled by Fur; however, whether Fur binds to FPI promoters directly has not been determined.

To determine if Fur binds to the promoters of the FPIs directly to repress FPI gene expression, we performed ChIP-Seq on cells grown in our standard iron-replete media to determine the location of Fur. We identified 21 Fur ChIP-Seq peaks; Fur ChIP-Seq peaks with associated genes (genes with translational start sites within either 100 bp upstream or 1 kb downstream from the maximum point of Fur enrichment) are listed in Table 3.2. As indicated in Table 3.2, expression of many of the genes associated with Fur ChIP-Seq peaks has been shown to change in response to low iron conditions (Deng et al., 2006). We did not detect any enrichment of Fur in the FPI, suggesting that Fur does not control expression of the FPI genes directly in response to iron (Appendix, Figure S1).

Table 3.2. Fur ChIP-Seq peaks and associated genes

Peak Number	Peak Maximum[1]	Peak Enrichment	Promoter-associated Peak[2]	Fur Box	Gene Distance[3]	Locus	Gene Name	Gene Product	Expression altered with iron limitation[4]
1	1771156	32.53	-	AAATGATAATG	94	FTL_1836	fslE	hypothetical protein	+
2	1765532	5.27	-	TAATGATAACG	893	FTL_1830	-	NADH dehydrogenase I subunit A	NA
					-8	FTL_1832	fslA	hypothetical protein	+
3	702485	2.2	-	GAATGATAAAG	-4	FTL_0712	-	hypothetical protein	+
4	1203636	2.19	-	GAATGATAACG	None	-	-	-	NA
5	490575	2.09	-	GAATGATAATG	None	-	-	-	NA
6	75226	1.72	-	GTTTGATAATG	682	FTL_0077	ribB	riboflavin synthase subunit alpha	+
7	1505709	1.69	-	GTTTGATAATG	514	FTL_1576	mutL	DNA mismatch repair protein	+
					171	FTL_1577	-	hypothetical protein	NA
8	140055	1.69	+	TAATGATAATT	159	FTL_0133	feoB	ferrous iron transport protein	+
9	492728	1.69	-	GAATGATAATG	897	FTL_0507	pyrE	orotate phosphoribosyltransferase	+
					630	FTL_0509	yjfH	tRNA/rRNA methyltransferase	+
10	400326	1.69	-	AAATGATAATG	914	FTL_0433	rrmJ	ribosomal large subunit methyltransferase J	+
11	1894397	1.65	+	AATAATTGATTATA	880	FTL_1966	trpE	anthranilate synthase component I	NA
					584	FTL_1967	-	trp operon repressor	NA
					252	FTL_R0051	tRNA-Asp1	Asp tRNA	NA
					172	FTL_R0052	tRNA-Thr1	Thr tRNA	NA
12	806879	1.64	-	AATGTATGAGAATG	39	FTL_0826	-	hypothetical protein, pseudogene	NA
13	1053271	1.64	-	GATGATTGATAATC	427	FTL_1108	pepB	cytosol aminopeptidase	+
14	60663	1.64	-	CATGTATGATAATT	None	-	-	-	NA
15	1444495	1.64	-	GCTGTTTGATAATA	687	FTL_1514	ybhR	ABC transporter permease	NA

Table 3.2. Fur ChIP-Seq peaks and associated genes, continued

16	553127	1.62	-	CTCGTATGAGAATG	None	-	-	-	NA
17	1860741	1.62	-	CATGTTTGATAATT	None	-	-	-	NA
18	498125	1.6	-	CATGTATGATAAGT	835	FTL_0514	-	hypothetical protein	+
					227	FTL_0515	-	ABC transporter substrate-binding protein	+
19	1269420	1.59	-	AATGTTTGAGAATG	865	FTL_1333	csdB	selenocysteine lyase	+
20	259025	1.56	-	CCAACATGATAATG	545	FTL_0273	gabP	glutamate/gamma-aminobutyrate anti-porter	+
21	72130	1.54	-	CATGTATGATAACT	451	FTL_0073	-	hypothetical protein	+

[1]Site of maximum ChIP-Seq peak enrichment

[2]"+", ChIP-Seq peak is promoter associated; "-", ChIP-Seq peak is not promoter associated

[3]Distance is measured as bp from the peak maximum to the annotated translational start site. "None", no genes identified with translational start sites within 100 bp upstream to 1 kb downstream of the site of peak maximum.

[4]As reported by Deng et al., 2006. "+", transcript levels increased; "-", transcript levels decreased; "NA", transcripts not affected or no gene examined.

The two most highly enriched Fur ChIP-Seq peaks are found at the *fsl* (or *fig*) operon, between the end of the *fur* gene and upstream of *fslA*, and between *fslD* and *fslE* (Figure 3.7). The *fsl* operon encodes a siderophore biosynthesis and uptake system used for iron uptake (Deng et al., 2006; Sullivan et al., 2006). Other Fur ChIP-Seq peaks are associated with genes coding for proteins involved in amino acid biosynthesis or other transport proteins, including an alternate iron uptake protein encoded by *feoB* (Thomas-Charles et al., 2013). Of the 21 Fur ChIP-Seq peaks, only two are found to intersect with promoter regions. This is consistent with the role of Fur as a repressor that binds to the promoters of Fur-regulated genes and occludes RNAP, thus preventing transcription under iron-replete conditions.

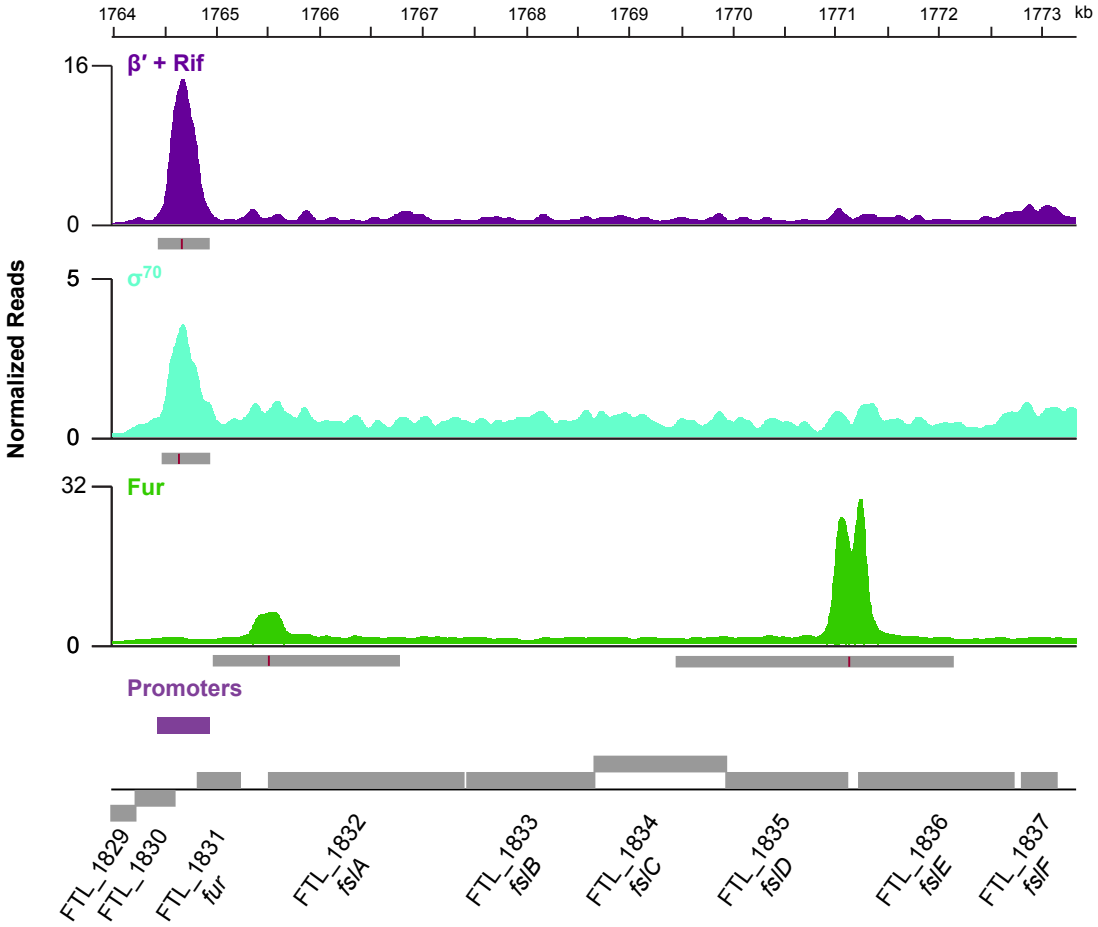
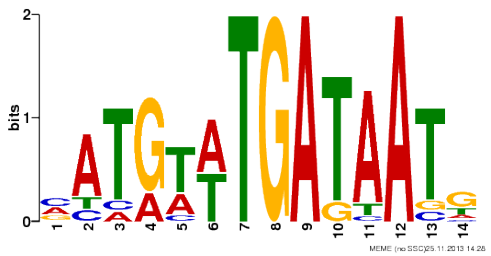


Figure 3.7. Regions of Fur enrichment are found at the *fsl* operon. Genomic position is indicated on the X-axis by *F. tularensis* LVS locus number; gray boxes represent genes, which are above the black line if they are encoded on the plus strand and below the black line if they are encoded on the minus strand. A representative track illustrating the density of normalized mapped sequencing reads after ChIP-Seq of each factor is depicted on the Y-axis, β′+ rif (purple), σ^{70} (aqua), and Fur (green). Areas of significantly enriched reads, peaks, are indicated by the horizontal gray boxes below the read density plot and sites of maximum enrichment are indicated by the red lines. Putative promoter regions, which are areas with significant enrichment of β′ + rif, σ^{32}, or σ^{70} association with the chromosome, are indicated by the purple boxes above the gene annotations.

The specific DNA sequence that Fur binds, the Fur box, has been described in *E. coli* and *Bacillus subtilis* as a 19-21 bp motif consisting of several repeats of a GATAAT hexamer, with the hexamer existing in several possible orientations (Baichoo and Helmann, 2002). Using MEME, we examined all of the sequences associated with the *F. tularensis* Fur ChIP-Seq peaks (Bailey and Elkan, 1994). We identified a 14 bp motif that contains elements of the Fur box GATAAT hexamer; we refer to the identified motif as the Ft-Fur box (Figure 3.8A and B). Although we have only found Fur associated with 21 regions, the Ft-Fur box sequence can be found 1,003 times in the LVS genome (motif sequences found with $p < 0.0001$; Grant et al., 2011). It seems reasonable that the likelihood of Fur associating with a particular Fur box is modulated by a number of factors, including the amount of Fur in the cell, the relative affinity Fur has for a particular sequence and the occupancy of other protein factors nearby. Consistent with the idea that Fur binding may be in part influenced by the affinity of Fur for a particular sequence, if we only search for a motif in 10 regions most enriched for Fur, a different consensus motif is identified (Figure 3.8C and D), which may be closer to the ideal Fur binding site. This motif associated with higher enrichment of Fur is 11 bp and contains one GATAAT hexamer.

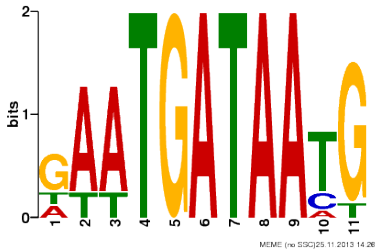
A.



B.

Sequence name[1]	Strand	Start	P-value		Site	
5-490575	+	259	7.43e-08	TGCGTATGAC	CATGAATGATAATG	TCACAGTCTT
14-60663	-	165	3.04e-07	CTTTTTGCTT	CATGTATGATAATT	AGCTGGGAAA
17-1860741	+	237	3.59e-07	CAAGGATTAG	CATGTTTGATAATT	GCACAAGAGG
12-806879	+	111	5.75e-07	TAGGGATAAA	AATGTATGAGAATG	GTCATTTTAT
19-1269420	-	224	6.75e-07	TTGATGAGAT	AATGTTTGAGAATG	AAAAGACAGC
6-75226	+	143	9.20e-07	AGGTTTTTTG	GTTGTTTGATAATG	AAAAAATATT
7-1505709	+	155	1.39e-06	TACCATCATC	AACGTTTGATAATG	GCACATAAAC
21-72130	-	155	2.62e-06	GATTCTTTAT	CATGTATGATAACT	TAGAAGAGCA
18-498125	-	295	2.62e-06	AGGTTTTGGG	CATGTATGATAAGT	GGTCCTATTT
13-1053271	-	263	2.91e-06	AGTAAACTCA	GATGATTGATAATC	ATAAGCTGCT
4-1203636	+	226	3.32e-06	ATTAACAATT	AATGAATGATAACG	TGTTACAGCA
15-1444495	-	195	4.53e-06	TAACCAACTT	GCTGTTTGATAATA	CTCGCGTGAG
2-1765532	-	170	6.70e-06	TTTTAAAATC	CCTACATGATAATG	ATAACGAATA
16-553127	+	315	8.21e-06	GGGTGAGAAA	CTCGTATGAGAATG	CTTTTGGCTA
10-400326	+	199	9.87e-06	CATTTGCCAT	GATGTTTGATCATT	GCTTAGAAGA
8-140055	-	138	2.13e-05	TATAATAATA	AATATTTGATAATA	ATTATCATTA
1-1771156	-	237	2.49e-05	TAGATATTAA	ATTAAATGATAATG	ATAATCAATA
9-492728	+	132	3.77e-05	TCTACAACGC	TAAGAATGATAATG	TACAAAAAAT
3-702485	+	94	4.02e-05	TAGTTTTTTG	GATGTTTGATTAGC	AGTTATATAC
20-259025	-	135	4.27e-05	TATCTGTGAG	CCAACATGATAATG	TAATCCTATT
11-1894397	-	210	1.25e-04	CTTCTGAAAA	AATAATTGATTATA	TAGAAAAAGA

C.



D.

Sequence name[1]	Strand	Start	P-value		Site	
9-492728	+	135	7.60e-07	ACAACGCTAA	GAATGATAATG	TACAAAAAAT
5-490575	+	262	7.60e-07	GTATGACCAT	GAATGATAATG	TCACAGTCTT
4-1203636	+	229	1.09e-06	AACAATTAAT	GAATGATAACG	TGTTACAGCA
10-400326	-	183	7.56e-06	ACATCATGGC	AAATGATAATG	TTTTCTACAA
3-702485	+	282	7.56e-06	GACTTTGGTA	GAATGATAAAG	TTCTCATTTG
1-1771156	-	237	7.56e-06	ATATTAAATT	AAATGATAATG	ATAATCAATA
2-1765532	-	164	9.08e-06	CCCTACATGA	TAATGATAACG	AATATCATTA
7-1505709	+	158	9.84e-06	CATCATCAAC	GTTTGATAATG	GCACATAAAC
6-75226	+	146	9.84e-06	TTTTTTGGTT	GTTTGATAATG	AAAAAATATT
8-140055	+	128	4.73e-05	GTAGATGTAA	TAATGATAATT	ATTATCAAAT

Figure 3.8. Defining the Fur box in *F. tularensis*. A. The sequence logo of the *F. tularensis* Fur box (Ft-Fur box), as identified by examining all 21 Fur ChIP-Seq peaks. **B.** The alignment of the putative Fur boxes found in all the sequences associated with Fur ChIP-Seq peaks. **C.** The sequence logo of the Fur box identified using the 10 most enriched Fur ChIP-Seq peaks. **D.** The alignment of putative Fur boxes found in the sequences associated with the 10 most enriched Fur ChIP-Seq peaks.

[1]Sequence name refers to the peak number, hyphenated with number of the chromosomal base pair that refers to the maximal point of Fur enrichment for the ChIP-Seq peak.

A sequence similar to the Fur box described in *E. coli* and *B. subtilis* has been identified within 100 bp of the annotated translational start sites for the *iglC* and *pdpB* genes. Despite the widespread presence of the Ft-Fur box, it was not detected within 1 kb of the annotated translational start site of the *pdpB* gene. The closest Ft-Fur box upstream of *iglC* is 566 bp from the annotated translational start site. However, the *iglABCD* genes are thought to consist of an operon, under the control of the *iglA* promoter (Appendix, Figure S1; Buchan et al., 2008). We do not find any evidence of an alternate promoter for the *iglC* gene, and there does not appear to be a Ft-Fur box within 1 kb of the *iglA* transcriptional start site. Thus, it seems unlikely that Fur would directly associate with FPI promoters to regulate gene expression.

Identification of the regulatory targets of OxyR and FTL_0040

Of the transcription factors we studied, many are, like Fur, found mostly at a few promoter or intergenic regions, suggesting they may regulate genes in a manner similar to canonical transcription activators or repressors. For each of the putative site-specific DNA binding proteins we examined, we have identified all the associated genes (genes which have translational start sites within either 100 bp upstream of or within 1kb downstream of the maximum point of enrichment of the factor) and proposed a putative regulon (see Appendix, Tables S3-S16). By identifying the direct targets of a virulence-associated transcription factor, we may be able to identify the gene(s) responsible for the transcription factor's contribution to virulence and reveal the regulatory hierarchy in the cell. We have specifically documented the virulence-associated genes found within the regulons of two factors, OxyR and the LysR-type transcription regulator FTL_0040.

In *E. coli*, the LysR-type transcription regulator OxyR typically functions as an activator and is responsible for initiating a cellular program in response to oxidative stress. In *F. tularensis*, the OxyR homolog has been implicated in virulence (Moule et al., 2010; Zheng et al., 2001). Specifically, a mutant of *F. novicida* containing a transposon insertion in *oxyR* was

attenuated for virulence in a *Drosophlia* model of infection (Moule et al., 2010). We found OxyR to be enriched at 27 genomic regions and identified 54 associated genes (Table 3.3). One of the genes associated with OxyR enrichment is *oxyR* itself, suggesting that in *F. tularensis*, as in *E. coli*, OxyR may regulate its own expression (Christman et al., 1989; Warne et al., 1990).

Table 3.3. OxyR ChIP-Seq peaks and associated genes

Peak Number	Peak Maximum[1]	Enrichment Factor	Promoter-associated[2]	Gene Distance[3]	Gene Locus	Gene Name	Citation linking gene to virulence	Gene Product
1	1648400	142.04	+	250	FTL_1717	*yaaA*	-	conserved protein YaaA
2	975430	58.94	+	48	FTL_1014	*oxyR*	(Moule et al., 2010)	oxidative stress transcriptional regulator
				50	FTL_1015	-	(Asare and Abu Kwaik, 2010; Guina et al., 2007; Wehrly et al., 2009)	AhpC/TSA family protein
				577	FTL_1016	-	(Meibom et al., 2009b; Wehrly et al., 2009)	short chain dehydrogenase
3	377297	51.9	+	843	FTL_0410	-	-	hypothetical protein
4	1436682	46.62	+	155	FTL_1504	*katG*	(Asare et al., 2010; Guina et al., 2007; Hazlett et al., 2008; Lindgren et al., 2007; Meibom et al., 2009b; Su et al., 2007; Wehrly et al., 2009)	peroxidase/catalase
5	343990	45.59	+	821	FTL_0370	-	(Kraemer et al., 2009)	arsenical restistance operon repressor
				361	FTL_0372	*gpsA*	(Kraemer et al., 2009)	NAD(P)H-dependent glycerol-3-phosphate dehydrogenase
6	1167578	15.37	+	353	FTL_1217	-	-	hypothetical protein
7	1335790	11.37	+	36	FTL_1407	*thrS*	-	threonyl-tRNA synthetase
				470	FTL_1408	-	-	chitin binding protein
8	178232	7.69	+	635	FTL_0173	*aroE*	(Moule et al., 2010)	shikimate 5-dehydrogenase
				42	FTL_0175	*rpmH*	-	50S ribosomal protein L34
				166	FTL_0176	*rnpA*	-	ribonuclease P protein component
				495	FTL_0177	-	(Asare et al., 2010)	hypothetical protein
				752	FTL_0178	*yidC*	(Asare et al., 2010)	inner-membrane protein
9	1421961	5.74	+	None	-	-	-	-
10	1306817	4.58	-	44	FTL_1374	*ostA2*	(Asare et al., 2010; Kraemer et al., 2009; Margolis et al., 2010)	organic solvent tolerance protein
11	1554413	4.16	+	97	FTL_1623	-	(Asare et al., 2010; Su et al., 2007)	hypothetical protein

Table 3.3. OxyR ChIP-Seq peaks and associated genes, continued

				175	FTL_1624	-	(Asare et al., 2010)	major facilitator transporter
12	1894310	3.95	+	793	FTL_1966	-	(Kadzhaev et al., 2009; Kraemer et al., 2009)	anthranilate synthase component I
				497	FTL_1967	-	-	trp operon repressor
				165	FTL_R0051	*tRNA-Asp1*	-	Asp tRNA
				85	FTL_R0052	*tRNA-Thr1*	-	Thr tRNA
13	338738	3.58	+	680	FTL_0362	-	-	pseudogene, hypothetical protein
				80	FTL_0363	-	-	psuedogene, LysR family transcriptional regulator
				59	FTL_0364	-	-	psuedogene, conserved hypothetical protein
				425	FTL_0365	-	-	pseudogene, conserved hypothetical protein
14	1333949	2.86	+	918	FTL_1404	*rplT*	(Su et al., 2007)	50S ribosomal protein L20
				685	FTL_1405	*rpmI*	-	50S ribosomal protein L35
				175	FTL_1406	*infC*	-	translation initiation factor IF-3
15	478974	2.54	-	103	FTL_0493	-	-	hypothetical protein
16	1642913	2.33	+	971	FTL_1709	-	-	hypothetical protein
				190	FTL_1710	*proQ*	(Asare et al., 2010; Weiss et al., 2007)	ProP osmoprotectant transporter, fragment
				-13	FTL_R0042	*tRNA-Met1*	-	Met tRNA
17	978368	2.08	-	29	FTL_1018	*serC*	-	phosphoserine aminotransferase
				291	FTL_1020	*isftu1*	-	transposase
18	1566067	1.88	-	900	FTL_1636	-	-	hypothetical protein
19	32121	1.83	+	232	FTL_0030	*carA*	(Ahlund et al., 2010; Asare and Abu Kwaik, 2010; Llewellyn et al., 2011; Qin and Mann, 2006; Schulert et al., 2009; Weiss et al., 2007)	carbamoyl phosphate synthase small subunit
				185	FTL_0031	-	-	acid phosphatase
20	424477	1.8	+	656	FTL_0448	-	-	M50B family metallopeptidase
				548	FTL_0449	*pigR*	(Brotcke and Monack, 2008; Buchan	hypothetical protein

Table 3.3. OxyR ChIP-Seq peaks and associated genes, continued

							et al., 2009; Charity et al., 2009)	
21	1546258	1.73	+	16	FTL_1616	*pckA*	(Asare et al., 2010)	phosphoenolpyruvate carboxykinase
				135	FTL_1617	*glnS*	-	glutaminyl-tRNA synthetase
22	223195	1.73	+	127	FTL_R0009	*tRNA-Glu1*	-	Glu tRNA
23	379908	1.65	+	64	FTL_0411	-	-	hypothetical protein
24	880014	1.65	+	366	FTL_0901	-	-	monooxygenase family protein
				202	FTL_0902	-	-	oxidoreductase
				-17	FTL_0903	*hflK*	(Asare et al., 2010; Su et al., 2007)	HflK-HflC membrane protein complex, HflK
25	996140	1.58	+	273	FTL_1039	-	-	hypothetical protein
				977	FTL_1040	-	-	hypothetical protein
				114	FTL_R0027	-	-	-
26	1414381	1.58	+	-49	FTL_R0040	*tRNA-Lys1*	-	Lys tRNA
27	918531	1.55	+	201	FTL_0948	-	-	Succinylglutamate desuccinylase / Aspartoacylase family protein
				90	FTL_0949	*prsA*	-	ribose-phosphate pyrophosphokinase

[1]Site of maximum ChIP-Seq peak enrichment.

[2]"+", ChIP-Seq peak is promoter associated; "-", ChIP-Seq peak is not promoter associated.

[3]Distance is measured as bp from the peak maximum to the annotated translational start site. "None", no genes identified with translational start sites within 100 bp upstream to 1 kb downstream of the site of peak maximum.

About half of the OxyR ChIP-Seq peaks, 13, are associated with a total of 19 genes previously implicated in virulence (Table 3.3). Only one of the virulence genes associated with OxyR ChIP-Seq peaks, *aroE*, was found in the same screen that originally identified OxyR as important for virulence (Moule et al., 2010). This raises the possibility that the attenuation phenotype of the *oxyR* mutant might be explained entirely through the effect of OxyR on *aroE* expression. It is also possible that the attenuated phenotype of the *oxyR* mutant might be explained by the effects of OxyR on any of the other 18 virulence genes it might control. Indeed, while OxyR is found associated with a promoter upstream of the *aroE* gene, there is another separate promoter region closer to the translational start site of *aroE* that OxyR is not associated with, which suggests that the OxyR-associated promoter may not be the promoter that controls *aroE* expression. The observed virulence defect of the OxyR mutant, then, may be related to the misregulation of one or more of the other 18 virulence-associated genes within the putative OxyR regulon. Together, this work suggests that OxyR coordinately regulates a number of virulence factors, including other transcription regulators that control virulence.

In *F. novicida*, a mutant containing a transposon insertion in the gene encoding the homolog of the LysR-type transcription regulator FTL_0040 was specifically attenuated for dissemination to the spleen in a mouse inhalation model of infection (Kraemer et al., 2009). In LVS, we identified 18 regions of FTL_0040 enrichment, 11 of which are found near translational start sites of genes (Table 3.4). The gene FTL_0040 is associated with the most highly enriched peak, a peak that is also promoter-associated, suggesting that FTL_0040 may regulate its own expression.

Table 3.4. FTL_0040 ChIP-Seq peaks and associated genes

Peak Number	Peak Maximum[1]	Peak Enrichment	Promoter-associated Peak[2]	Gene Distance[3]	Locus	Gene Name	Citation linking gene to virulence	Gene Product
				641	FTL_0038	*emrA2*	-	HlyD family secretion protein
1	41227	141.55	+	145	FTL_0039	-	(Asare et al., 2010)	hypothetical membrane protein
				-19	FTL_0040	-	(Kraemer et al., 2009)	LysR family transcriptional regulator
				417	FTL_0742	-	(Moule et al., 2010)	LysR family transcriptional regulator
2	732761	6.65	+	140	FTL_0743	-	-	oxidoreductase, short-chain dehydrogenase family protein
				996	FTL_0744	*sbcB*	(Kraemer et al., 2009)	exodeoxyribonuclease I
				202	FTL_0061	-	(Asare and Abu Kwaik, 2010)	hypothetical protein
3	61557	5.26	+	999	FTL_0063		-	major facilitator superfamily (MFS) transport protein
				693	FTL_1932	-	-	pseudogene, dienelactone hydrolase family protein
4	1864487	4.93	-	153	FTL_1933	-	-	pseudogene, dienelactone hydrolase family protein
				55	FTL_1934		-	permease of ABC transporter
5	1833177		+	149	FTL_1902	-	(Kraemer et al., 2009)	conserved hypothetical membrane protein
6	139931	3.65	+	35	FTL_0133	*feoB*	(Ahlund et al., 2010; Asare and Abu Kwaik, 2010; Thomas-Charles et al., 2013)	ferrous iron transport protein
7	1061935	2.93	-	None	-	-	-	-
8	762152	2.78	-	None	-	-	-	-
9	1039986	2.64	-	None	-	-	-	-
10	1571718	2.59	-	None	-	-	-	-
11	1822778	2.43	-	110	FTL_1889	-	-	3-isopropylmalate dehydratase large subunit
				168	FTL_1891	*isftu1*	-	transposase

Table 3.4. FTL_0040 ChIP-Seq peaks and associated genes, continued

12	586966	2.42	-	137	FTL_0600	*wbtH*	-	asparagine synthase
13	46475	2.06	-	None	-	-	-	-
14	1745171	2.04	-	None	-	-	-	-
15	37134	1.73	+	423	FTL_0034	-	-	hypothetical protein
				114	FTL_0035	-	-	hypothetical protein
16	50621	1.61	-	86	FTL_0048	*tyrA*	(Kraemer et al., 2009)	prephenate dehydrogenase.
17	1496256	1.58	-	122	FTL_1567	*araJ*	(Asare and Abu Kwaik, 2010)	major facilitator superfamily (MFS) transport protein
				-11	FTL_1568	-	-	LysR family transcriptional regulator
				946	FTL_1569	*gph*	(Kraemer et al., 2009)	phosphoglycolate phosphatase
18	345700	1.53	-	None	-	-	-	-

[1]Site of maximum ChIP-Seq peak enrichment

[2]"+", ChIP-Seq peak is promoter associated; "-", ChIP-Seq peak is not promoter associated

[3]Distance is measured as bp from the peak maximum to the annotated translational start site. "None", no genes identified with translational start sites within 100 bp upstream to 1 kb downstream of the site of peak maximum.

Of the 11 FTL_0040 ChIP-seq peaks, 7 are associated with genes implicated previously as virulence factors through a variety of studies (Table 3.4). Four genes associated with FTL_0040 enrichment peaks, *sbcB*, FTL_1902, *tyrA*, and *gph*, were also identified in the screen that originally implicated FTL_0040 in virulence (Kraemer et al., 2009). FTL_1902 encodes a hypothetical protein predicted to localize to the membrane, and a FTL_1902 mutant was, like a FTL_0040 mutant, found to be specifically attenuated for dissemination to the mouse spleen (Kraemer et al., 2009). This suggests that the specific virulence defect observed with the FTL_0040 mutant might be completely explained by the loss of FTL_1902 expression in the FTL_0040 mutant. By determining the targets of FTL_0400 by ChIP-Seq we may have identified a pathway directly involved in signaling the production of factors required for infection of a specific niche.

A model of the virulence regulatory network in *F. tularensis*

The ChIP-Seq analysis of the putative OxyR and FTL_0040 regulons reveals that both transcription factors are found at the promoters of genes encoding other transcription factors. Our data also indicate that one function of PmrA may be to control gene expression indirectly through its direct effect on the expression of *pigR*. Given these relationships between transcription factors, we asked if we could identify a network or hierarchy of transcription factor regulation.

We examined the regulon of each transcription factor as determined by ChIP-Seq and asked if any regulon members encoded a transcription factor. Because PigR is found at essentially all promoters, we excluded its potential effect on any transcription factor apart from the documented effect of PigR on its own promoter (Charity et al., 2009). We also excluded from our analyses the putative NAP IclR, as previously reported microarray results suggest that IclR does not influence the expression of any genes encoding a transcription factor (Mortensen et al., 2010). We have suggested that PmrA controls gene expression, at least in part, by

promoting the expression of *pigR*, and so we only included the documented effect of PmrA on *pigR* expression (Mohapatra et al., 2007; Sammons-Jackson et al., 2008).

Figure 3.9 illustrates a model of the virulence regulatory network in *F. tularensis* revealing the connections between different virulence regulators. OxyR is found at the promoters of the genes encoding PigR and ArsR, both of which have been implicated in virulence, as well as the FTL_1967 locus, which has not been implicated in virulence. FTL_0040 appears to regulate an entire section of the network. FTL_0040 is found at the promoter of another transcription factor demonstrated to be important for virulence, FTL_0742, as well as at the promoter of FTL_1568, which has not been associated with virulence. However, FTL_1568 is found at the promoter regions of five other transcription factors, all of which have been previously implicated in virulence. The net result of these interactions suggests that FTL_0040 may be at the top of a regulatory hierarchy, regulating the expression of six other transcription factors important for *F. tularensis* pathogenesis.

The network depicted in Figure 3.9 also reveals that multiple transcription factors implicated in virulence may regulate their own expression. For one factor, we only detected enrichment of the factor at its own putative promoter (ArsR, see Appendix, Table S7). In this instance it is unclear whether the factor controls the expression of itself in addition to virulence genes present in the same operon, or whether the factor binds other regions involved in virulence that are not detected in our analyses. Generating a model of the regulation of transcription factors, such as in Figure 3.9, provides insight into the complexity of gene regulation in *F. tularensis* and provides us with the opportunity to generate new hypotheses regarding virulence factor regulation.

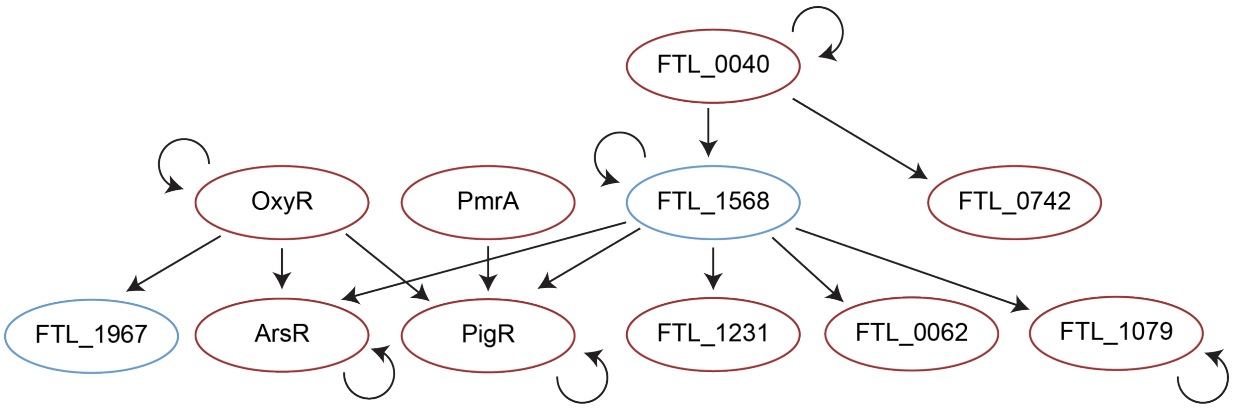


Figure 3.9. The network of virulence regulators in *F. tularensis* LVS. Circles indicate transcription factors; those in red have previously been implicated in virulence while those in blue have not. Arrows indicate the presence of a transcription factor at the putative promoter of the gene encoding another transcription factor.

Discussion

We have performed ChIP-Seq on 30 transcription factors in *F. tularensis* LVS, including all those implicated in the control of virulence gene expression. By determining the location of each transcription factor, we have been able to predict potential regulons under the direct control of each factor. For the predicted OxyR and FTL_0040 regulons, we have determined which regulon members have previously been implicated in virulence, allowing us to offer possible explanations for how each of these regulatory factors directly contributes to virulence. We have presented evidence that the positive effect of PmrA on the expression of genes located on the FPI may be indirect, through PmrA promoting *pigR* expression. Additionally, PmrA may be functioning along with FTL_1568 and IclR as a NAP. We have described the putative regulon of the ferric uptake regulation protein, Fur, and demonstrated that it may not directly control the expression of FPI-encoded genes. Finally, we have been able to generate a model of the regulatory network that controls virulence in *F. tularensis*, suggesting a regulatory hierarchy.

We identified 3 transcription factors which may act in part as NAPs, specifically PmrA, FTL_1568, and IclR. The ChIP-Seq peaks identified for the putative NAPs vary widely with respect to fold enrichment, suggesting that some sites may be high affinity sites while others may be weaker in affinity for the particular factor. However, the relationship between fold enrichment and affinity of a particular transcription factor for a given site is not well characterized. There are multiple factors that determine the extent to which a transcription factor associates with a particular region. Degree of occupancy of a particular site might be influenced by the affinity the factor has for a particular site, by competition with other factors for the available site, and by nucleoid structure; relative enrichment values in our identification of ChIP-Seq peaks do not necessarily correlate to relative binding affinity.

Many of the ChIP-Seq peaks for PmrA, FTL_1568, and IclR are poorly-enriched sites, that is, are enriched less than 2-fold (24%, 30%, and 21% of ChIP-Seq peaks for PmrA,

FTL_1568, and IclR, respectively). Sites with widespread weak enrichment for a particular transcription factor have been reported in eukaryotes (Farnham, 2009; Galagan et al., 2013b; MacQuarrie et al., 2011) and prokaryotes (Chumsakul et al., 2013; Galagan et al., 2013a; 2013b). In *Mycobacterium tuberculosis*, at least a subset of weak association sites appear to be functional, in that they affect the regulation of the surrounding gene(s) (Galagan et al., 2013a). It is possible that these poorly enriched sites are lower in affinity for the particular transcription factor and would be more highly enriched in the context of greater transcription factor abundance, as has been demonstrated in *M. tuberculosis* (Galagan et al., 2013a). Identification of weakly enriched sites could also result from proteins that are merely found in close proximity to specific regions of DNA, such as regions where DNA may be tightly compacted in a specific structure.

Enrichment of PmrA, IclR, and FTL_1568 is, at their respective most enriched ChIP-Seq peak, relatively high in comparison to the other factors this study has examined. It might be argued then that all transcription factors occupy a range of high and low enrichment sites and the only reason we see many poorly enriched sites for these three factors is because they are more easily detected by ChIP-Seq than others we have examined. However, we have not necessarily found that having ChIP-Seq peaks with very high enrichment always correlates to having many peaks with low enrichment. One LysR-type transcription regulator in particular, FTL_1222, is found to be essentially as enriched as FTL_1568 at its highest ChIP-Seq peak (1,126-fold versus 1,223-fold enriched, respectively), and more enriched than the highest PmrA ChIP-Seq peak (1,126-fold versus 536-fold). However, rather than being found at hundreds of locations across the genome, FTL_1222 is only associated with three. This suggests that the correlation between high enrichment and ChIP-Seq peak abundance is not absolute and suggests that the large numbers of peaks identified are not artifacts of the ChIP-Seq protocol.

Potential artifacts of ChIP-Seq experiments are a concern, as a recent paper demonstrating that certain highly-transcribed regions of the eukaryotic *Saccharomyces*

cerevisiae genome are easily identified by ChIP-Seq, even when using a heterologous control protein that is not known to bind DNA (Teytelman et al., 2013). However, our data seem to be close to ideal in terms of avoiding artifacts resulting from highly-transcribed regions. In this study, we compare DNA specifically immunoprecipitated from cell lysates containing the epitope-tagged transcription factor of interest (IP DNA) to DNA non-specifically immunoprecipitated from cell lysates lacking epitope-tagged protein (mock IP DNA). Both DNA samples undergo some selection using the same antibody-decorated beads. Similar to the results of Teytelman et al., we find that even in the mock IP DNA, immunoprecipitation results in enrichment of particular genomic regions (data not shown, Brencic et al., 2009), which we can effectively subtract from further analyses. But in the analysis of *S. cerevisiae*, certain regions were still found to be consistently enriched when a protein was immunoprecipitated, even a heterologous, non-DNA-binding protein such as GFP. Therefore, the authors suggest comparing peaks found using a similar heterologous control to peaks specifically identified using the transcription factor of interest, or to at least compare many datasets. The work presented here consists of 30 ChIP-Seq experiments of different factors performed in triplicate using the same protocol, all of which are compared to the mock IP experiment. Thus, we have generated a large dataset in which to detect potential artifacts from highly-expressed regions. While it is notable that one factor in particular, FTL_1231, is mostly weakly enriched at the highly-transcribed tRNA genes, we do not see a specific pattern of enrichment common to ChIP-Seq of all transcription factors. This suggests that ChIP-Seq of highly transcribed regions is not a significant problem in our dataset.

It is entirely possible, however, that detecting artifactual ChIP-Seq enrichment at highly-expressed loci is dependent on a certain level of protein expression. In our experiments, transcription factors are expressed from their native promoter on the chromosome. The level of native expression is likely to be different for each factor. In fact, ChIP-Seq of several transcription factors, FTL_1216, FTL_0844, and FTL_1634, did not identify any peaks. This may

reflect a lack of protein expression under the conditions used; consistent with this, when generating strains that would produce the epitope-tagged proteins encoded by FTL_1216, FTL_0844 and FTL_1634, we could not detect epitope-tagged protein by Western blot.

A technical limitation of our approach to ChIP-Seq lies in our use of epitope-tagged factors to determine the location of DNA-associated proteins. It is possible that the addition of the epitope tag interferes with the function of one or more factors, potentially reducing their ability to bind DNA. However, we were able to identify ChIP-Seq peaks for 90% of the factors we performed ChIP-Seq upon, implying that the addition of the epitope tag did not completely abolish the association of the factor with DNA for most factors.

We compared the location of ChIP-Seq peaks for PmrA and IclR to reported transcriptomic data in order to correlate transcription factor binding with potential function (Mortensen et al., 2010; Sammons-Jackson et al., 2008). In the case of both PmrA and IclR, we found poor correlation between reported regulated genes and genes associated with a ChIP-Seq peak. Specifically, of the 17 genes whose expression is altered in the absence of IclR (Mortensen et al., 2010), only 7 are found with translational start sites within 1kb of an IclR ChIP-Seq peak maximum. PmrA is reported to control the expression of 155 genes in LVS (Sammons-Jackson et al., 2008), but only 35 were found to have a translational start site within 1 kb of a PmrA ChIP-Seq peak maximum. These findings are consistent with the idea that IclR and PmrA do not always function as canonical transcription factors and may exert some of their effects on gene expression indirectly.

A study of PmrA in *F. novicida* has also reported transcriptomic data and there is poor correlation between the genes reported to be regulated by PmrA in LVS and *F. novicida*; of the 65 PmrA-regulated genes reported in *F. novicida* (Mohapatra et al., 2007), only 5 overlap with the 155 PmrA-regulated genes in LVS (Sammons-Jackson et al., 2008). Genes positively regulated by PmrA in both analyses include three genes located on the FPI and *pigR*; the one gene shown to be negatively regulated by PmrA in both studies is the gene encoding the

hypothetical protein FTL_0702, which is a gene associated with the most highly enriched PmrA ChIP-Seq peak. The apparently poor overlap in regulons between the two strains raises the possibility that there may be some strain-specific differences in the role of PmrA on gene expression.

It should be noted that, in our comparative analysis of trancriptiomic data and ChIP-Seq data, there are a number of caveats. The lack of correlation between identified regulated genes in previous studies and identified ChIP-Seq peaks from this work is also consistent with differences in a number of experiment-specific possibilities. These include growth media specific or growth phase specific effects, or differences between detection limits for microarray versus ChIP-Seq experiments.

However, it is also a possibility that many of the individual ChIP-Seq peaks we have identified for PmrA, FTL_1568, and IclR, all of which exhibit widespread genomic association in both promoter and non-promoter locations, do not result in detectable regulatory consequences on nearby genes. Rather than to regulate gene expression, these sites may be important to regulate chromosome structure and reflect a NAP function for PmrA, FTL_1568, and IclR. In fact, LysR-type transcription regulators such as FTL_1568 are thought to function, at least in part, to activate or repress transcription by binding and bending DNA, which would be consistent with a role both in transcription regulation and chromosome organization. The binding and bending of DNA by transcription factors may have additional consequences; it has been suggested that transcription factor binding may influence the accessibility of DNA to both mutagenic agents and repair enzymes (Warnecke et al., 2012). Thus, transcription factor binding within coding regions, by proteins such as PmrA, FTL_1568, and IclR may have biological consequences for the evolution of a gene (Warnecke et al., 2012).

Work performed in *F. novicida* has resulted in a model for how PmrA controls the expression of virulence genes present on the FPI (Bell et al., 2010). In this model, PmrA is proposed to bind directly to promoter regions on the FPI and function as a transcription activator

by interacting with the RNAP-associated MglA-SspA complex (Bell et al., 2010). Using ChIP-Seq in LVS, we failed to detect PmrA at either the *iglA* or *pdpA* promoters, suggesting that PmrA does not influence the expression of FPI genes directly in this organism. While it is possible that our inability to detect PmrA at the *iglA* and *pdpA* promoters simply reflects an issue with the sensitivity of our detection method, we did reproducibly detect PmrA at many other regions with enrichment values between 1.5 and 536-fold. Nevertheless, we did detect PmrA at the *pigR* promoter using ChIP-Seq and PmrA has been shown to positively regulate *pigR* expression (Mohapatra et al., 2007; Sammons-Jackson et al., 2008). Furthermore, our ChIP-Seq studies identified MglA, SspA, and PigR associated with FPI promoters, which suggests that the role of MglA, SspA, and PigR on FPI gene expression is direct. Our findings with ChIP-Seq are more consistent with an alternative model in which PmrA influences the expression of genes on the FPI indirectly through an effect on expression of *pigR* (Meibom et al., 2009a; Figure 3.10). Through the regulation of the PigR transcription factor, PmrA may indirectly control the entire PigR regulon.

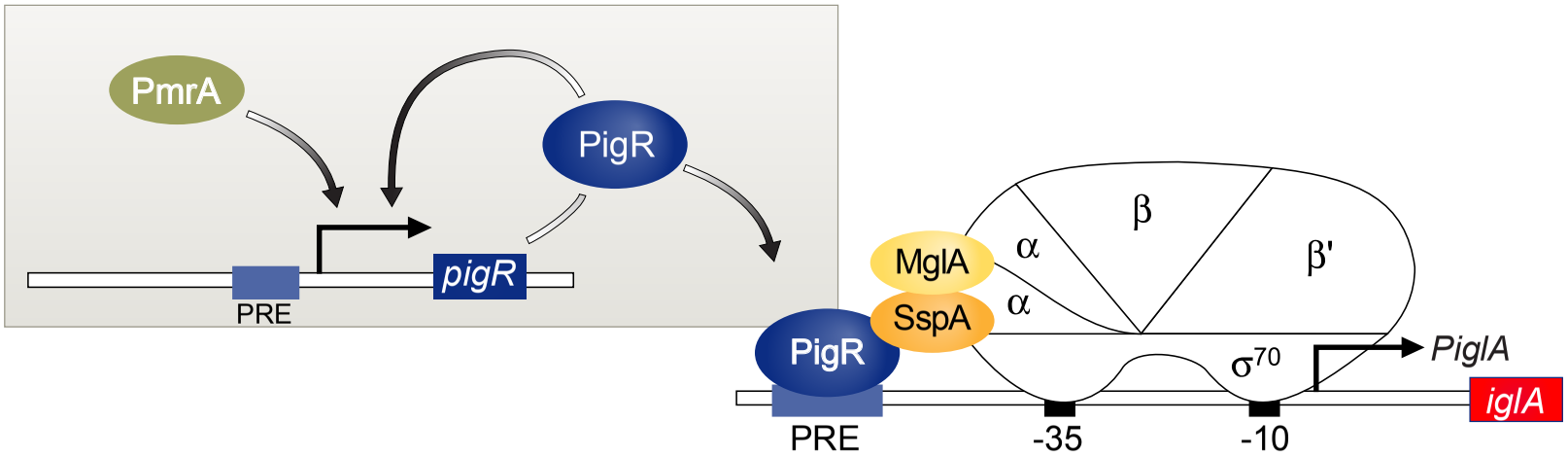


Figure 3.10. PmrA promotes expression from the *pigR* gene, indirectly regulating FPI gene expression. The transcription factor PmrA directly associates with the *pigR* promoter to regulate *pigR* gene expression. The resulting PigR protein interacts directly with the RNAP-associated MglA-SspA complex and stabilizes the binding of RNAP at specific target promoters containing a PigR response element (PRE, Chapter 2) such as the FPI-encoded *iglA* promoter.

The host intracellular environment is thought to have a limited amount of easily accessible iron (Caza and Kronstad, 2013; Schaible and Kaufmann, 2004), and iron limitation promotes expression of the FPI-encoded virulence genes in *F. tularensis* (Buchan et al., 2008; 2009; Deng et al., 2006; Lenco et al., 2007). However, the mechanism behind this regulation is unclear. Because Fur is considered the major iron-responsive transcription factor, it is an attractive candidate for regulation of genes affected by iron abundance and putative Fur boxes were identified at FPI promoters (Deng et al., 2006). However, several studies have suggested that Fur does not directly regulate FPI gene expression (Buchan et al., 2008; 2009). Our results are consistent with these data, as we did not detect Fur association with FPI promoters. Additionally, our studies identified a Fur box sequence specific to *F. tularensis*, the Ft-Fur box, which is not identified near FPI promoter regions. Thus, it seems likely that another mechanism is responsible for promoting FPI gene expression in low iron conditions. In *E. coli*, SpoT appears to promote ppGpp formation under conditions of iron limitation (Vinella et al., 2005). In LVS, SpoT is required for FPI gene expression as well as intramacrophage growth and virulence (Charity et al., 2009). It is therefore possible that SpoT is the factor that, in response to low iron, promotes FPI gene expression in *F. tularensis* by promoting ppGpp formation.

While this point is implicit, it should be reiterated that transcription factors are considered virulence factors because they control the production of virulence factors. The LysR-type transcription regulators OxyR and FTL_0040 have been identified as virulence factors (Kraemer et al., 2009; Moule et al., 2010) and this work elucidates the virulence factors they are likely to regulate. While the identification for OxyR and FTl_0040 as virulence factors only reflects that they are necessary to regulate at least one virulence factor, both have putative regulons that contain multiple factors linked to virulence. This work suggests that both OxyR and FTL_0040 are important to coordinate a transcriptional program, rather than a single gene, important for virulence.

The OxyR regulon, based on work in other organisms, is generally thought to include genes required for the detoxification of reactive oxygen or nitrogen species (reviewed in Imlay, 2013). The genes implicated by ChIP-Seq as members of the LVS OxyR regulon appears to be similar to other OxyR homolog regulons (Wei et al., 2012; Zheng et al., 2001). Specifically, OxyR is found at the promoter of *katG*, which encodes catalase, and at the promoter of a gene encoding an AhpC family protein. Both of these proteins are commonly found under the control of OxyR, as they are used to detoxify hydrogen peroxide. The most enriched OxyR ChIP-Seq peak is at the promoter region of a gene annotated as encoding a hypothetical protein, FTL_1717. Upon further examination, it appears that this gene encodes the *F. tularensis* LVS homolog of the *E. coli* YaaA protein and so has been referred to as such in Table 3.3. In *E. coli*, *yaaA* is part of the OxyR regulon and its protein product has been shown to be important for lowering intracellular iron levels, although its mechanism of action is still unknown (Liu et al., 2011; Zheng et al., 2001). There are a few differences between the OxyR regulon in *F. tularensis* and other organisms; in *E. coli*, OxyR binds the *fur* promoter to directly regulate Fur expression (Zheng et al., 1999), but we do not detect enrichment of OxyR at the Fur promoter. Some differences are possibly due to the lack of *F. tularensis* homologs of OxyR regulon members found in other bacteria, such as the lack of a *F. tularensis* homolog for *dps*, which, in other bacteria, encodes a gene important to scavenge iron (reviewed in Imlay, 2013).

The studies of OxyR and FTL_0040 also highlight another important point regarding the information to be gleaned from this body of work. Each transcription factor linked to virulence was identified in a specific screen using a specific model for *F. tularensis* pathogenesis. As *F. tularensis* successfully proceeds through an infection, it must adapt and respond to a number of different environments in a variety of different cell types, organs, and even hosts. Many of these screens identify the same virulence factors, which are interesting because they seem to be universal to infection of all cells, organs, and hosts. Yet tropisms in infection, that is, preferences in infection sites or specific niches, can reveal important biological information about both the

pathogen and the host. At least some of the transcription factors in this study may be dispensable for virulence in a number of models, but have been found to be critical in at least one. This suggests that there are biologically relevant pathways required in, and possibly unique to, particular models of infection. By identifying the direct targets of transcription factors critical for pathogenesis within the context of different infections, we can start to identify these cell-, organ-, or host-specific pathways through which *F. tularensis* causes disease and learn more about specific infection tropisms.

The genes located on the FPI are essential for intracellular survival and growth and are required for virulence (Bröms et al., 2010). We have replicated the result that FPI gene expression is reduced in cells lacking PigR (Chapter 2, Table 2.2, Brotcke and Monack, 2008; Charity et al., 2009) and have found PigR at the FPI promoters (Figure 3.3), indicating that PigR directly promotes FPI gene expression. However, *pigR* expression may be subject to regulation by a number of factors. It is known that *pigR* is regulated by its own protein product and by MglA and SspA (Brotcke et al., 2008; Charity et al., 2009). It also appears that PmrA is a direct regulator of *pigR* expression (Figure 3.3; Mohapatra et al., 2007; Sammons-Jackson et al., 2008). The presence of OxyR, FTL_1568, and IclR are all detected upstream of the *pigR* promoter along with MglA, SspA, PigR, and PmrA. Additionally, expression of *pigR* appears to be subject to control by the small molecule ppGpp, which seems to promote, either directly or indirectly, the interaction between PigR and the MglA-SspA complex. Thus, regulation of *pigR* expression appears to be a critical step in regulating FPI gene expression.

Ultimately our data have led to a model for the network of transcription factor regulation in *F. tularensis* (Figure 3.9). The model of transcription regulation we have proposed highlights the interconnected relationships that exist between transcription regulators. This illustrates an important advantage of using ChIP-Seq to identify targets of regulation, in comparison to a transcriptomic approach. Generally, transcriptomic approaches compare cells lacking a protein to cells containing the protein of interest. This is a powerful approach, but if removal of one

transcription factor impacts the production of another transcription factor, it can be difficult to distinguish direct effects from indirect effects. An example of the ambiguity inherent to transcriptomic data is provided by PmrA. While microarray data are consistent both with PmrA directly regulating FPI gene expression and with PmrA indirectly regulating FPI gene expression through PigR, the PmrA ChIP-Seq data suggests that PmrA indirectly affects FPI gene expression through PigR.

It should be noted that we did not find much evidence of functional redundancy, i.e. significant overlap of operons, within the regulons of LysR family members or between other transcription factors. There are genes with promoters that intersect with the ChIP-Seq peaks of multiple family members, such as the FTL_1568 promoter, at which FTL_0040 is found with FTL_1568 itself. This could be indicative of co-regulation where these two members of the LysR family function together to control the expression of a particular gene. It is also possible that they recognize identical or overlapping DNA sequences and compete with one another for the available site(s). In this latter case, our observations would reflect the existence of a subset of cells with one factor associated with the promoter region, and another subset of cells with the other factor associated.

We found a number of transcription factors at their own promoter; of 20 factors (those for which we detected ChIP-Seq peaks, excluding RNAP subunits, the elongation factor GreA, MglA, and SspA) 9, or 45%, are detected near their putative promoter region. Autoregulation is a common feature of LysR-type transcription regulators, and we detected evidence of this for three of the seven LysR family members. We additionally found evidence of autoregulation for three proteins that are characterized in other species as repressors: LexA, HipB, and ArsR. PigR is found at its own promoter, and has previously been shown to regulate its own expression (Brotcke and Monack, 2008; Charity et al., 2009). The remaining proteins that are potentially autoregulatory are the two hypothetical transcription factors FTL_1079 and FTL_1222, both of which contain predicted helix-turn-helix motifs.

By determining the location of many transcription factors across the genome, we have gained insight into gene regulation in *F. tularensis* in a number of ways. This work has suggested alternate models for gene regulation of specific virulence factors critical for *F. tularensis* pathogenesis. Additionally, these data have led us to suggest that several proteins may play a role in chromosomal organization, which may open new and interesting lines of inquiry. Finally, while attempting to parse how transcription factors influence gene expression, we have gained new insights into how these factors are connected and the regulation to which they themselves are subject.

Materials and Methods

Plasmids, strains, and growth conditions

F. tularensis subsp. *holarctica* LVS was grown at 37°C in either Mueller Hinton (MH) broth (Difco) supplemented with glucose (0.1%), ferric pyrophosphate (0.025%), and Isovitalex (2%), or on cysteine heart agar (Difco) medium supplemented with 1% hemoglobin solution (VWR); when appropriate, kanamycin was used for selection at 5 µg/ml. *E. coli* strain XL1-blue (Stratagene) was used for plasmid construction and, when appropriate, kanamycin was used to select for resistance at 50 µg/ml.

Integration vectors, strain construction, Southern blotting and immunoblots

Integration constructs and strains were generated and confirmed as in Chapter 2. For genes whose putative operons may be interrupted by plasmid integration, a plasmid that promotes transcription downstream from the integrated plasmid was generated, as described in Chapter 2. This strategy was necessary to add the DNA encoding the epitope tag to both the *sspA* and *pmrA* genes.

Chromatin Immunoprecipitation (ChIP), Illumina Library Preparation, ChIP-Seq data analysis

ChIP was performed in biological triplicate (excepting β′ + rifampicin, OxyR, and LVS Δ*pigR* pF-PigR-V, which were performed in duplicate and σ^{70}, which was performed in quadruplicate) as described in Chapter 2. Library preparation for Illumina sequencing and subsequent data analysis were performed as described in Chapter 2.

MEME

To identify a common motif in Fur ChIP-Seq peaks, the 300 bp region surrounding the site of maximum Fur enrichment for each Fur ChIP-Seq peak was identified. The first Fur motif (Figure 3.8A and B) was identified by searching all of the Fur ChIP-Seq peaks for a common

motif using MEME; the second Fur motif (Figure 3.8C and D) was identified by searching only the ten most enriched Fur ChIP-Seq peaks using MEME (Bailey and Elkan, 1994). FIMO (Grant et al., 2011) was subsequently used to identify the occurrences of each motif in the *F. tularensis* LVS genome.

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Chapter 4

Summary and Future Directions

Summary

Francisella tularensis is the causative agent of the disease tularemia. As an intracellular pathogen, *F. tularensis* must be able to survive inside the host cell. One of the first genetic loci found to be critical for intracellular survival was named macrophage growth locus A, or *mglA* (Baron and Nano, 1998). A stringent starvation protein A (SspA) family member, MglA has since been found to be a transcription regulator essential to activate the transcription of genes critical for virulence encoded on the *Francisella* pathogenicity island (FPI) (Lauriano et al., 2004). Recent work has demonstrated that MglA functions as a heteromer with its homolog, SspA, in concert with another protein, PigR, to regulate gene expression (Brotcke and Monack, 2008; Charity et al., 2009).

Our laboratory previously proposed a model to explain how MglA, SspA, and PigR regulate a common set of genes. According to this model, PigR, a putative DNA-binding protein, binds to a specific DNA sequence at target promoters. At these locations, PigR would interact directly with the RNA polymerase (RNAP)-associated MglA-SspA complex to stabilize RNAP at the promoter and increase transcription of specific target genes (Charity et al., 2009).

Between our work and other studies, we are continuing to gain insight into the mechanisms by which MglA, SspA, and PigR function to regulate virulence gene expression. However, while a large number of other *F. tularensis* transcription factors have been implicated in virulence, very little is known about their role in regulating virulence factors. Transcription networks and hierarchies have been uncovered that govern the production of virulence factors in many other bacteria, but none have been described in *F. tularensis*.

Elucidating how virulence factors are coordinately regulated, by all transcription factors, will allow further insight into the mechanisms by which *F. tularensis* causes pathogenesis. Additionally, while we have proposed a model for the manner in which MglA, SspA, and PigR function to regulate gene expression, several features of this model had yet to be experimentally tested. These include that PigR is a site-specific DNA binding protein that is only found at target

promoters, and that MglA, SspA, and RNAP should be found together at target promoters with PigR.

In order to both answer specific questions regarding the mechanisms by which MglA, SspA, and PigR regulate virulence gene expression, as well to determine the direct targets of the other virulence-associated transcription regulators in *F. tularensis*, I performed chromatin immunoprecipitation followed by high-thoughput sequencing (ChIP-Seq) on a total of 31 *F. tularensis* transcription factors, which includes all of the transcription factors previously implicated in virulence and 70% of the total number of transcription factors thought to be present in *F. tularensis* LVS. This allowed me to identify all of the genomic regions associated with each transcription factor examined. As a result, we have revised our model of virulence gene regulation by MglA, SspA, and PigR, and have identified the putative direct targets for all of the transcription factors analyzed. In addition, these studies have led to several other hypotheses regarding virulence-associated transcription factors, including that several, specifically PmrA, IclR, and the LysR-type transcription regulator FTL_1568, may function as nucleoid-associated proteins (NAPs). I have made a connection between several virulence-associated transcription factors and specific genes previously proposed to be important for virulence, and finally, have presented a model describing the network of transcription factors controlling virulence in *F. tularensis*.

As described in Chapter 2, I detected MglA, SspA, and PigR at virtually every promoter region across the genome. This suggests that if PigR is a site-specific DNA-binding protein, the PigR-specific site would have to be present at all promoter regions. In this case, we hypothesized that PigR may bind the promoter-specific site regardless of the presence of the MglA-SspA complex. However, I did not detect PigR at promoter regions in the absence of MglA. This indicates that PigR requires MglA (and thus presumably, the MglA-SspA complex) to associate with promoter regions.

Since MglA, SspA, and PigR are found together at virtually all promoter regions and appear to control the same set of genes (Brotcke and Monack, 2008; Charity et al., 2009), it would be logical to suggest that these three proteins function together and control exactly the same genes. However, I observed a difference between cells lacking MglA in comparison with cells lacking PigR; as previously reported, cells lacking MglA had a slow growth phenotype (Charity et al., 2007; Honn et al., 2012), but cells lacking PigR grew similarly to wild-type cells. I therefore reasoned that there might be some differences between the genes that PigR and MglA regulate, and that MglA may be important in regulating genes critical to maintaining normal growth.

To determine if there are differences between the regulons of MglA and PigR, I assessed the transcript levels of a subset of *F. tularensis* genes, including genes reported to be positively or negatively regulated by MglA as well as those not known to be regulated by MglA, SspA, or PigR. I found that despite the concordance in localization as determined by ChIP-Seq, there are critical differences between the functions of MglA, SspA, and PigR. In association with MglA, PigR functions as a positive regulator. However, MglA also appears to function to repress gene expression independently of PigR.

In order to determine if there are any sequences specific to promoters that are responsive to PigR, I searched for a conserved motif upstream of PigR-regulated genes. I found a particular 7 bp sequence in the PigR-regulated promoters examined, which, in promoters with identified transcription start sites, is 6-8 bp from the predicted -35 element. This suggests that the conserved sequence, which we tentatively call the PigR response element (PRE), may be found at all PigR-regulated promoters and may allow PigR to function to positively regulate the promoter. There are a number of ways in which recognition of this motif could occur, including that PigR could directly interact with this element, or that the binding of PigR to the RNAP-associated MglA-SspA complex could promote the interaction between some other portion of RNAP, or the MglA-SspA complex itself, with the PRE.

From these results, we hypothesize that the role of MglA and SspA in virulence gene expression may be principally to serve as a point of contact for PigR to influence RNAP activity and positively regulate a specific set of genes. This hypothesis is consistent with a number of pieces of data, specifically (i) the direct interaction between PigR and the MglA-SspA complex, (ii) the requirement of MglA for PigR to associate with promoter regions, (iii) the positive regulation by PigR of a subset of the MglA regulon, and (iv) the similar virulence phenotypes of cells lacking MglA, SspA, or PigR.

Results from the transcriptomic data in Chapter 2 suggest that MglA, but not PigR, may play a role in negatively regulating genes, specifically, those with σ^{32}-controlled promoters. A possible explanation for this might be that the MglA-SspA complex promotes formation of σ^{70}-containing RNAP holoenzyme but not formation of the σ^{32}-containing holoenzyme. By influencing the competition between σ factors for core RNAP, the MglA-SspA complex would effectively decrease transcription from σ^{32}-dependent promoters and potentially even increase transcription from σ^{70}-dependent promoters. In other organisms, it is known that SspA homologs influence the transcription of genes controlled by specific σ factors (De Reuse and Taha, 1997; Hansen et al., 2003; Yin et al., 2013). It is possible that in these other organisms and in *F. tularensis*, SspA functions to influence gene expression by altering σ factor competition.

As described in Chapter 3, I used ChIP-Seq to identify putative regulons for many other transcription factors in *F. tularensis*. The majority of transcription factors in *F. tularensis* have been implicated in virulence, and I examined each of these, along with the entire family of annotated LysR-type transcription regulators.

The transcription factor PmrA has been demonstrated to positively regulate genes on the FPI (Mohapatra et al., 2007; Sammons-Jackson et al., 2008), yet I did not detect PmrA at key promoters on the FPI. However, I was able to detect PmrA at the *pigR* promoter and previous transcriptomic analyses indicated that PmrA positively controls *pigR* expression (Mohapatra et al., 2007; Sammons-Jackson et al., 2008). This suggests that *pigR* is a direct target of PmrA.

Since PigR is present at promoters on the FPI and has been shown to influence FPI gene expression, it follows that PmrA may positively regulate FPI gene expression indirectly, by promoting *pigR* expression.

The identification of PmrA, IclR, and the LysR-type transcription regulator FTL_1568 at many genomic regions, both promoter and non-promoter regions, suggests that these factors may not always act as canonical transcription regulators. Rather, the abundance and location of the ChIP-Seq peaks of PmrA, IclR, and FTL_1568 suggests that they may play a role in chromosomal organization as NAPs. As transcription factors continue to be examined on a genome-wide level by ChIP-Seq, it is becoming increasingly recognized that they may be found at non-canonical locations. At least some of these sites of association may be attributed to particular factors acting, not only as canonical transcription factors, but also as NAPs (Dillon and Dorman, 2010). A role for transcription factors in chromosomal organization is perhaps not surprising, given that many factors can recognize both specific and degenerate motifs and, in binding the DNA, can alter the physical orientation of the DNA molecule within the cell. Additionally, *F. tularensis* is only thought to encode one well-described NAP, the HU homolog HupB. Given the lack of annotated proteins with NAP function in *F. tularensis*, identifying several within this work would seem to be a reasonable expectation.

I investigated the location of the iron-dependent repressor Fur, not because it has explicitly been demonstrated to be important for virulence, but because FPI gene expression has been shown to increase under conditions of limited iron (Buchan et al., 2008; Deng et al., 2006; Lenco et al., 2005); increased gene expression under iron limitation is a hallmark of de-repression by Fur. Previous studies implied that Fur may be responsible for the increase in FPI gene expression under low iron conditions and suggested the specific locations with which Fur might associate to influence FPI gene expression (Deng et al., 2006; Lenco et al., 2005). However, my results do not indicate that Fur is found at the FPI, suggesting that Fur does not directly associate with the FPI. Additionally, my analysis of Fur location suggests a *F. tularensis*-

specific variation of the motif with which Fur associates. This specific motif is not found near FPI promoters. I do find Fur at the *fsl* operon, which has been shown to be repressed by Fur under conditions of high iron and has been identified as an important virulence factor in a number of screens. Thus, while Fur does not appear to directly influence FPI gene expression, I confirmed that it does appear to act directly to regulate another described virulence locus, the *fsl* operon.

In order to determine why OxyR and FTL_0040 are important for virulence, I examined their putative regulons to identify genes previously implicated as encoding virulence factors. I found that both OxyR and FTL_0040 appear to regulate multiple virulence-associated genes, suggesting that, rather than regulating one specific virulence factor, they are responsible for the coordinate regulation of multiple factors involved in pathogenesis.

We reasoned that, since infection and survival in a host cell is a complex multi-step process, there is likely to be significant regulation of transcription factors themselves. In fact, I found many transcription factors at the promoters of genes encoding other transcription factors. I examined the putative regulons of each factor for other transcription factors and was able to discern a network of potential regulation among the transcription factors. This network model suggests that several factors are ultimately responsible for regulating the expression of not only a number of transcription factors, but consequently, a large number of putative virulence factors.

My studies, focused on examining the location of transcription factors in association with the genome, provide us with more than the putative regulons for each virulence-associated transcription factor in *F. tularensis*. By determining the locations at which MglA, SspA and PigR are found, we have been able to revise our model suggesting how these transcription factors regulate virulence gene expression. For specific transcription factors, we have been able to suggest alternate functions. And we have gained insight into the architecture of virulence control in *F. tularensis*; we have shed light on the coordinate regulation of genes implicated in virulence, as well as the regulation of the genes that encode the transcription factors themselves.

Together, these data have provided a framework for understanding how virulence may be regulated in *F. tularensis*.

Future Directions

Chapters 2 and 3 describe the identification of the genomic locations for thirty-one *F. tularensis* transcription factors. These studies have yielded a wealth of information with respect to transcription regulation in *F. tularensis*. However, the mechanism by which MglA, SspA, and PigR regulate virulence gene expression remains unclear. Additionally, while we have proposed putative regulons for all of the studied transcription factors, we do not know to what extent or manner these factors affect their putative regulons. Accordingly, this section includes a discussion of potential future experiments to explore these, and other, questions.

Does the MglA-SspA complex influence σ factor competition?

The SspA homolog found in several other organisms seems to function, as a dimer, by promoting expression from genes controlled by particular σ factors. In *Escherichia coli*, SspA was found to associate with RNAP containing σ^{70} but not core RNAP or σ^{S}-containing RNAP. Our data suggest that in *F. tularensis*, MglA may function to inhibit transcription from σ^{32}-regulated promoters. However, our data do not address whether or not the change in σ factor competition promoted by MglA and SspA might lead to an increase in transcription from σ^{70}-dependent promoters. We hypothesize that the presence of the MglA-SspA complex promotes the formation of RNAP containing σ^{70}, and in doing so, decreases the amount of core RNAP available to associate with σ^{32}. Thus, in the absence of MglA, the competition between σ factors for the limited pool of RNAP is altered; without MglA, more core RNAP would be available to interact with σ^{32}, allowing for increased transcription from σ^{32}-regulated promoters.

To more directly test if the increased expression from σ^{32}-regulated promoters results from an increase in σ^{32} binding to σ^{32}-regulated promoters, one could quantify the amount of σ^{32}-regulated promoter DNA immunoprecipitated by σ^{32} in wild-type cells in comparison to cells lacking MglA. This would indicate that there is a difference in the amount of σ^{32} available in the

cell to associate with σ^{32}-regulated promoters. However, this would not entirely rule out the possibility that MglA directly binds to, and represses, σ^{32}-regulated promoters.

In order to determine if loss of MglA has a direct effect on the amount of RNAP holoenzyme containing a particular σ factor, one could immunoprecipitate RNAP and determine the relative amounts of σ^{32} and σ^{70} associated with RNAP in cells with MglA compared to cells lacking MglA. This could be done using the strain in which the β′ subunit of RNAP is synthesized with a tandem affinity purification (TAP) tag, which was previously used to identify MglA, SspA, and σ^{70} associated with RNAP in *F. tularensis* (Charity et al., 2009). Immunoprecipitating more σ^{32}-associated with RNAP in the absence of MglA than in the presence of MglA would be consistent with the MglA-SspA complex affecting σ factor competition, although it would not directly suggest a mechanism by which the complex promotes formation of the σ^{70}-containing holoenzyme. In this case, further genetic and biochemical assays would be necessary to elucidate the mechanism through which the MglA-SspA complex functions. Note that it is not currently known which subunit (or subunits) of RNAP is (are) contacted by the MglA-SspA complex. Biochemical or genetic (bacterial two-hybrid) studies could in principle determine where the MglA-SspA complex contacts RNAP, and could reveal whether the MglA-SspA complex might specifically interact with the σ^{70} subunit of RNAP.

A corollary of our hypothesis, that the MglA-SspA complex may function to promote the formation of RNAP containing σ^{70}, is that we would not expect the MglA-SspA complex to be associated with RNAP containing σ^{32}. To directly test if the MglA-SspA complex is associated with RNAP containing σ^{32}, one could immunoprecipitate σ^{32}-containing RNAP and examine the associated proteins for the presence of either MglA or SspA. This could be done by generating a strain in which σ^{32} is synthesized with a TAP tag. The absence of MglA and SspA associated with σ^{32}-containing RNAP would further suggest that the MglA-SspA complex specifically interacts with σ^{70}-containing RNAP. A similar immunoprecipitation could be performed using σ^{70} synthesized with a TAP tag as a control for the ability to detect MglA and SspA; if similar ratios

of RNAP subunits are associated with both σ factors, but MglA and SspA are not associated with σ^{32}, this would provide evidence that the MglA-SspA complex only associates with σ^{70}-containing RNAP. However, one would have to demonstrate that the cells that produce TAP-tagged σ factors contain the same amount of MglA and SspA.

Does the PigR response element (PRE) confer regulation by PigR?

The identification of a specific sequence in PigR-regulated promoters, 6-8 bp from the -35 element, suggests that this motif, the PRE, may be an element which allows the activity of a promoter to respond to the presence of PigR. One could test this by introducing a reporter gene, such as *lacZ*, under the control of a PigR-regulated promoter, into strains with and without PigR. We expect that, using a PigR-regulated promoter with an intact PRE, *lacZ* expression in cells containing PigR would be significantly higher than in cells that did not contain PigR. However, if significant changes are made to alter the PRE, we expect that reporter gene expression would only be significantly affected in cells that produce PigR. One could also test this by introducing the PRE into a non-PigR regulated promoter and seeing whether the modified promoter now becomes responsive to PigR.

It is important to reiterate that, in the case that the PRE does function as a PigR response element, this does not necessarily mean that PigR must bind or contact the PRE directly to promote gene expression at promoters with a PRE. It is possible that the presence of PigR affects RNAP to allow a specific domain of RNAP to contact the PRE and promote transcription. The C-terminal domain of the α subunit of RNAP (αCTD) has been found to contact a region upstream of the -35 element, and could be appropriately positioned to contact the PRE.

An indirect measure of determining if the PRE allows control by PigR would be to determine if all of the PigR-regulated promoters contain a PRE. However, the PRE motif, as currently defined, is only 7 bp, of which only 3 bp are completely conserved; the PRE is likely to

be found frequently throughout the genome. In order to firmly establish that both the PRE sequence and location, with respect to transcription start site, are conserved at all PigR-regulated promoters, one would have to identify all the transcription start sites throughout the genome, which could be done using RNA-Seq.

Can we correlate transcription factor location with transcription factor regulation?

In this study, we have identified putative direct targets, or regulons for each transcription factor examined. However, these data do not indicate if the association of a transcription factor with a particular location results in gene regulation. In fact, genes found to be regulated by PmrA and IclR in previously published transcriptomic studies do not correlate well to genes found near the ChIP-Seq peak for each factor. It is entirely possible that the presence of a factor at a particular location does not have any regulatory consequences; it is equally possible that the lack of overlap between regulated genes and genes associated with a ChIP-Seq peak is due to technical differences between experiments. Thus, the natural complement of ChIP-Seq analysis is transcriptomic analysis; identifying those genes whose expression changes in the absence of the transcription factor would allow us to suggest connections between the presence of a specific factor at a specific region and any associated changes in gene expression. Transcriptomic analyses of factors could be done at the whole-genome level by RNA-Seq using strains that lack a particular transcription factor.

Analysis of the regulon of select transcription factors by analyzing transcript abundance would also allow us to test and refine our model of regulation by transcription regulators. While we currently know which transcription factors may be directly influencing other transcription factors, the ChIP-Seq peaks identified are agnostic with respect to positive or negative regulation. A much greater understanding of the virulence control network could be gained by ascertaining the potential regulatory consequences of a ChIP-Seq peak.

Could we assess transcription factor location at the whole-genome level?

In this study, we have determined the transcription factor locations for a larger proportion of total encoded transcription factors than any known study to date, reporting ChIP-Seq peaks for 70% of all known transcription factors in *Francisella tularensis*. If we consider that RNAP is a single transcription factor, we have assessed approximately 83% of transcription factors and only 11 of the total 40 remain to be examined. By performing ChIP-Seq on the remaining 11 factors, we could obtain a whole-genome view of transcription factor association in a single organism. This may allow us to determine how many promoters in an organism are actually regulated by transcription factors other than RNAP. It also may provide further insight into any additional proteins important for chromosomal organization.

Could performing ChIP-Seq *in vivo* or under alternate conditions identify additional targets?

We have essentially taken a snapshot of transcription regulation within *F. tularensis* in a particular condition at a particular stage of growth. While FPI-encoded virulence genes are expressed under these conditions, our growth conditions are unlikely to accurately mimic the range of environments which *F. tularensis* is exposed to, nor are they likely to mimic the conditions found in the intracellular environment, when survival of *F. tularensis* relies on the appropriate regulation of virulence factor production. In fact, *in vitro* growth media for *F. tularensis* have been reported which appear to be more similar to an *in vivo* environment than the media which we use (Hazlett et al., 2008). Under alternate conditions, it is likely that some of the transcription factors we have studied associate with different genomic regions and different target genes. If we examined transcription factor location in the context of other growth conditions, or in an *in vivo* model system (such as within macrophage), we may gain significant insights into how particular transcription factors are regulated and function to regulate target genes and virulence factors.

Performing ChIP-Seq using cells from an infection model, or *in vivo* ChIP-Seq, would yield the direct targets of virulence-associated transcription factors while they are in the processes of regulating pathogenesis. Transcriptomic analyses have successfully been performed in macrophage cell lines (Bent et al., 2013; Wehrly et al., 2009), which may be the most practical option with the technology we have currently. It should be noted that the significant technical challenge of *in vivo* ChIP-Seq would still be obtaining sufficient immunoprecipitated DNA for sequencing. Each *F. tularensis* cell can have hundreds of thousands of copies of a particular transcript, but each cell contains only a single chromosome. At best, each cell can only yield a single fragment for each site of transcription factor association. However, detection methods continue to improve and *in vivo* ChIP-Seq would give us a greater understanding of how virulence factors are regulated in an intracellular context, yielding insight into the extent to which transcription factor location differs between *in vitro* and *in vivo* conditions.

For three factors, we failed to identify any ChIP-Seq peaks. There are a number of possible reasons for the lack of ChIP-Seq peak detection. It is possible that the addition of the epitope tag significantly interfered with the function or DNA binding of these factors. Alternately, it is possible that under the conditions of our experiments, the three factors were not made. Because each transcription factor examined is under the control of its native promoter and the conditions under which the native promoter is active have not been explored, it is possible that the protein levels of some transcription factors were very low or nonexistent. Consistent with this idea, detecting epitope-tagged protein was unsuccessful for several transcription factors; the addition of DNA encoding the epitope tag had to be confirmed at the DNA level. One protein which did not ChIP-Seq successfully, encoded by the locus FTL_1216, has recently been found to be highly upregulated at the transcript level in macrophages (Bent et al., 2013). Finding this factor highly upregulated in an infection model reiterates its importance in virulence and suggests that, rather than the protein not having any direct targets, we have been unsuccessful

at identifying them. In order to successfully define the regions of association for the factors that do not yet have any associated ChIP-Seq peaks, we could perform ChIP-Seq on cells grown in alternate conditions or *in vivo*, after validating protein production.

Overproduction of transcription factors is another strategy we could utilize to gain insight into the potential targets of a particular transcription factor, specifically for those factors that do not yet have any associated ChIP-Seq peaks. This approach has been successful in the past (Davies et al., 2011), and, if protein association with its direct targets relies mostly on affinity of the protein for a particular DNA site, an abundance of protein could potentially allow us to saturate the factor's target sites. However, if a given transcription factor requires the presence of another factor to bind or associate with the DNA, or can only associate with regions that are not bound by another highly expressed transcription factor, we may not achieve a complete understanding of its target sites.

Concluding statement

A significant proportion of transcription factors encoded by *F. tularensis* are utilized to coordinately control the regulation of putative virulence factors, and these transcription factors themselves are subject to a network of regulation. MglA, SspA, and PigR, factors critical for intracelluar growth and survival, are found at essentially all promoters. The MglA-SspA complex may influence gene expression by playing a role in promoting formation of σ^{70}-containing RNAP and also by providing a point of contact for PigR with the RNAP complex. PigR, in concert with MglA and SspA, acts as a positive regulator of gene expression and appears to be regulated by a number of factors. PigR, together with MglA and SspA, regulates *pigR* expression, as does PmrA and the small molecule ppGpp; OxyR, IclR, and FTL_1568 are all also detectable at the *pigR* promoter by ChIP-Seq, although the potential regulatory consequences of these associations is currently unknown. The PmrA, FTL_1568, and IclR proteins are found associated with many locations that are non-canonical for transcription factors, potentially indicating an alternate role for these factors in chromosomal organization. The LysR-type transcription regulators OxyR and FTL_0040 appear to coordinately regulate a number of putative virulence factors. Together, these studies allow for the generation of a model that suggests how transcription factors important for virulence are themselves controlled, which features a significant amount of potential autoregulation. The data presented here yield insights into the manner by which *F. tularensis* coordinately regulates virulence at the level of transcription.

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Appendix

Supporting Information

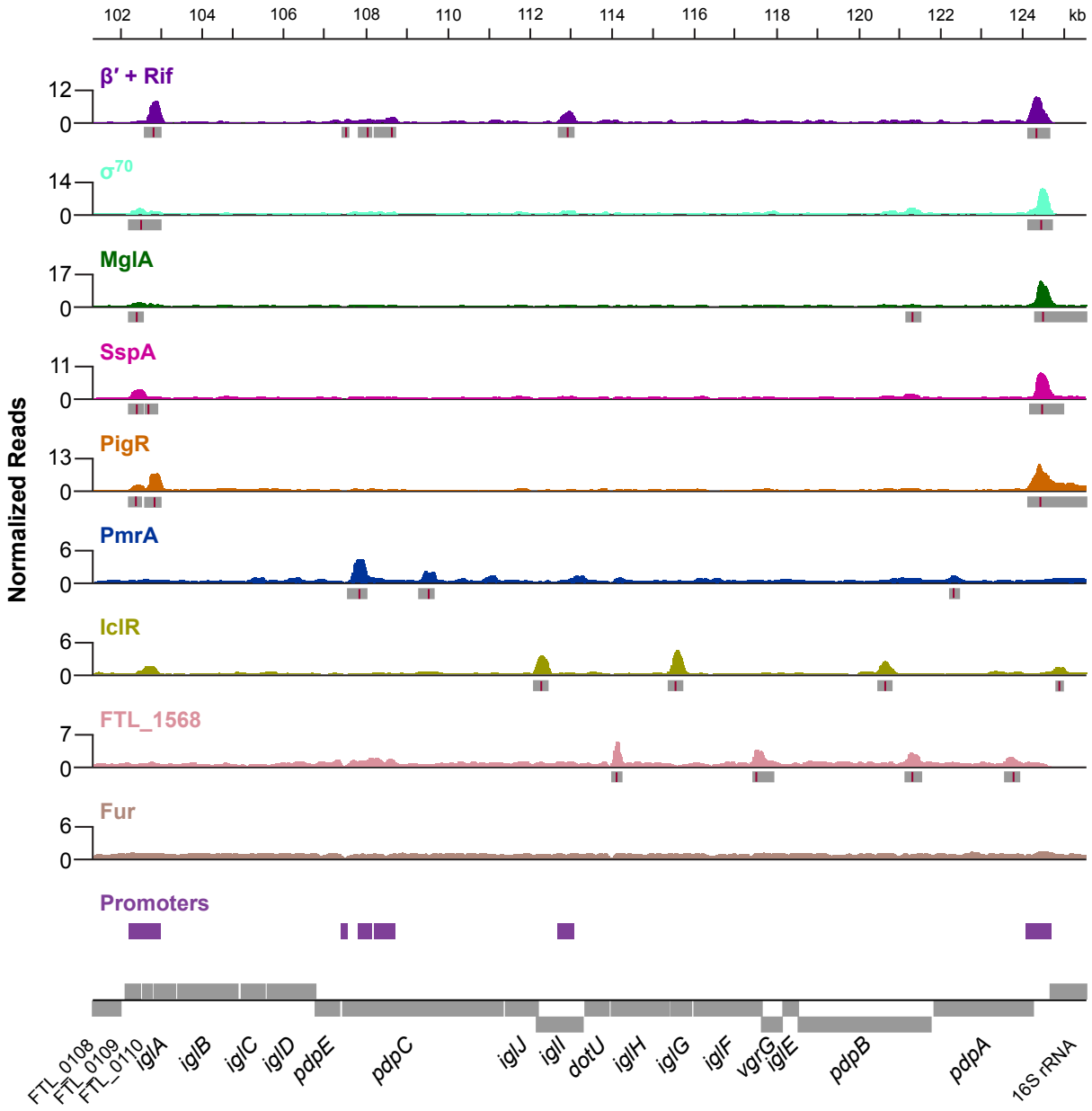

Figure S1. Proteins associated with the FPI. Genomic position is indicated on the X-axis by *F. tularensis* LVS locus number; gray boxes represent genes, which are above the black line if they are encoded on the plus strand and below the black line if they are encoded on the minus strand. Representative tracks illustrating the density of normalized mapped sequencing reads after ChIP-Seq of β′ + rif, σ^{70}, MglA, SspA, PigR, PmrA, IclR, FTL_1568, and Fur are depicted on the Y-axis. Areas of significantly enriched reads, referred to here as peaks, are indicated by the horizontal gray boxes below the read density plot and sites of maximum enrichment are indicated by the red lines. Putative promoter regions, which are areas with significant enrichment of β′ + rif, σ^{32}, or σ^{70} association with the chromosome, are indicated by the purple horizontal boxes above the genomic position markers. PmrA is not enriched at either the *iglA* or *pdpA* promoter, unlike β’ + rif, σ^{70}, MglA, SspA, and PigR. IclR and FTL_1568 are largely found in non-promoter-associated regions.

Table S1. Transcription factors in *F. tularensis* LVS

LVS[1]	U112[2]	SCHU S4[3]	Gene Name	Product
FTL_0034	-	FTT1659		hypothetical protein
FTL_0040	FTN_0031	FTT1652c	-	LysR family transcriptional regulator
FTL_0062	FTN_0101	FTT1684	-	transcription regulator
FTL_0261	FTN_0264	FTT0350	*rpoA1*	DNA-directed RNA polymerase, α subunit
FTL_0370	FTN_0395	FTT0868c	*arsR*	arsenical resistance operon repressor
FTL_0415	-	FTT1307c	-	hypothetical protein
FTL_0428	FTN_0434	FTT0908	*parB*	chromosome partition protein B
FTL_0449	FTN_0480	FTT0383	*pigR*	transcription factor PigR
FTL_0552	FTN_1465	FTT1557c	*pmrA*	two-component response regulator
FTL_0610	FTN_1416	FTT1446	*rho*	transcription termination factor Rho
FTL_0616	FTN_1412	FTT_1442c	*rpoA2*	DNA-directed RNA polymerase, α2 subunit
FTL_0632	FTN_1393	FTT_1426	-	ArsR family transcriptional regulator
FTL_0643	FTN_1384	FTT1418c	*nusB*	transcription antitermination factor NusB
FTL_0662	FTN_1363	FTT1401	*lexA*	prophage repressor protein
FTL_0689	FTN_1274	FTT1255c	-	transcriptional regulator *araC* family protein
FTL_0742	FTN_1179	FTT1202	-	LysR family transcriptional regulator
FTL_0844	FTN_1099	FTT1119	-	LysR transcriptional regulator family protein
FTL_0851	FTN_1092	FTT1112c	*rpoH*	RNA polymerase σ^{32} factor
FTL_0895	FTN_1054	FTT0627	*hupB*	Histone-like protein HU form B
FTL_1014	FTN_0959	FTT0556c	*oxyR*	oxidative stress transcriptional regulator
FTL_1050	FTN_0913	FTT1035c	*rpoD*	RNA polymerase σ^{70} factor
FTL_1079	FTN_0889	-	-	helix-turn-helix family protein
FTL_1126	FTN_0795	FTT1076	*hipB*	transcriptional regulator / hipB?
FTL_1176	FTN_1300	FTT1285c	-	LysR transcriptional regulator family protein
FTL_1185	FTN_1290	FTT1275	*mglA*	macrophage growth locus, subunit A
FTL_1193	FTN_1282	FTT1267	-	LysR transcriptional regulator family protein
FTL_1216	FTN_0866	FTT0985	-	hypothetical protein FTL_1216
FTL_1222	FTN_0858	FTT0977c	-	hypothetical protein FTL_1222
FTL_1231	FTN_0850	FTT0970	-	hypothetical protein FTL_1231
FTL_1276	FTN_0811	FTT0477	*birA*	Bifunctional protein BirA
FTL_1293	FTN_1534	FTT0359	-	hypothetical protein FTL_1293
FTL_1364	FTN_0720	FTT0748	*iclR*	hypothetical protein FTL_1364
FTL_1474	FTN_0665	FTT1313c	*greA*	transcriptional elongation factor
FTL_1533	FTN_0613	FTT0703	*rpoZ*	DNA-directed RNA polymerase, ω subunit
FTL_1568	FTN_0583	FTT0492c	-	LysR family transcriptional regulator
FTL_1606	FTN_0549	FTT0458	*sspA*	stringent starvation protein A, regulator of transcription

Table S1. Transcription factors in *F. tularensis* LVS, continued

FTL_1631	-	-	-	hypothetical protein
FTL_1634	-	FTT1594	-	LysR family transcriptional regulator
FTL_1743	FTN_1567	FTT0145	*rpoC*	DNA-directed RNA polymerase, β' subunit
FTL_1744	FTN_1568	FTT0144	*rpoB*	DNA-directed RNA polymerase, β subunit
FTL_1749	FTN_1573	FTT0139	*nusG*	transcription antitermination protein NusG
FTL_1810	FTN_1661	FTT0049	*nusA*	transcription elongation factor NusA
FTL_1831	FTN_1681	FTT0030c	*fur*	ferric uptake regulation protein
FTL_1967	FTN_1779	FTT1803c	*trpR*	*trp* operon repressor

[1]Gene locus number in *F. tularensis* subsp. *holarctica* strain LVS

[2]Gene locus number in *F. tularensis* subsp. *novicida* strain U112

[3]Gene locus number in *F. tularensis* subsp. *tularensis* strain SCHU S4

Table S2. Putative promoter regions identified in *F. tularensis* LVS

Start	End
1	339
7942	8338
8600	9045
10748	11651
11845	12188
19094	19714
23025	23370
24170	24809
24926	25623
31789	32439
34105	34392
35695	37036
39468	39778
40647	41418
43389	43682
47556	48046
55912	56290
56788	57162
58960	59377
60861	61192
61319	61724
65926	66391
67249	68017
75915	76021
76709	76816
78626	79113
79943	80255
81141	81474
83928	84498
91780	92198
92546	92984
95189	95457
98274	98473
99396	99767
102223	103009
107450	107571
107826	108163
108214	108730
112693	113081
124133	124730
129577	129942
133883	134330
136750	137704
139794	140180
141784	143324
143759	144355
148687	149029
149556	149929
152853	153186
153315	153690
158224	158923
162129	162949
166014	166474
168413	168728
169341	169765
172933	173301
176613	176993
177460	177828
178020	178591
178607	179126
181242	181985
190201	190692
192181	192567
199044	199335
207094	207416
213655	213967
222833	223600
224258	224596
224792	225274
230656	231072
247708	247989
248600	250221
254735	255023
261051	261502
265780	266260
267837	268195
271797	272166
272589	272915
273254	273591

Table S2. Putative promoter regions identified in *F. tularensis* LVS, continued

275587	275889
276135	276444
277816	278525
278649	278967
280492	280961
281260	281721
286869	287212
289225	289981
293009	293610
293835	294242
303936	304273
309053	309477
311506	311929
314555	314999
317074	317372
317980	318184
326905	327231
327604	328293
329242	329554
332243	332625
333647	333962
334626	335238
335821	336165
337624	337906
338554	338870
340453	340736
344038	344465
347952	348288
349519	349980
350908	351322
351996	352459
353865	354128
356708	357189
361081	361407
363272	363709
371106	371861
372054	372437
373443	373807
376974	377421
377950	378381
379663	380079
380734	381012
384137	384428
388116	388988
389175	390104
390873	391243
392275	392599
399294	399708
403193	403568
404129	404662
405386	405845
406174	406587
408034	408303
409032	409142
413903	414414
415098	415752
420579	420927
421628	422089
422641	423465
423777	424090
424194	425158
425786	426466
428093	428419
431452	431608
432457	433394
435502	435905
440294	440741
441122	441430
444056	444818
445448	446070
449968	450360
452645	453086
454877	455276
456749	457022
465650	465993
468009	468320
470285	470631
476004	476302
476737	477129
487819	488091
491699	492120
495346	495874

Table S2. Putative promoter regions identified in *F. tularensis* LVS, continued

500608	500984
501162	502479
504507	504765
504911	505308
509462	509735
513539	514057
519934	520315
521222	521581
526474	527227
527755	528144
530764	531270
533778	534506
537122	537387
545701	545843
547976	548278
549272	549671
555729	556266
557034	557503
567042	567416
568797	569199
569293	569624
572270	572613
577203	577499
582221	582530
584637	585136
602423	602873
605821	606241
607364	608505
609593	609943
612455	612766
627802	628561
629739	630074
632946	633482
634321	634665
636269	636967
637710	638082
641600	642078
643615	643828
644799	645287
646342	646650
662668	662924
670860	671114
676478	676837
679132	679399
680642	681244
685723	686042
692179	692310
693492	693891
695408	695615
698904	699341
701518	701623
709592	710001
713540	713957
714321	714804
719210	719580
721445	721870
723648	723927
725840	726364
728181	728506
729677	730020
732692	732974
733570	733861
736979	737202
738771	739029
740182	740458
746613	746873
747754	748086
749955	750385
759642	759969
761743	761962
771198	771578
774604	774945
776079	776347
777429	777731
780280	780624
781457	781832
784607	784977
786105	786476
787863	788176
788562	789074
793172	793475
797803	798086

Table S2. Putative promoter regions identified in *F. tularensis* LVS, continued

801771	801876
803217	803506
805023	805557
810078	810491
811431	811724
811854	812005
816002	816446
817697	818031
818842	819218
822585	822671
823757	824088
824190	824499
826432	826731
827971	828735
829410	829804
831788	832260
838283	838754
840327	840595
843815	844276
845448	845706
848016	848495
849710	849966
852179	852536
854109	854378
858849	859224
859965	860470
865473	865852
867909	868474
873821	874311
874813	875196
876711	877044
879711	880116
883001	883303
883777	884014
886231	886572
889491	889901
890324	890614
893616	894102
894326	894820
897376	897754
898910	899233
899538	899714
904624	905191
906804	906910
907673	907986
914342	914778
918321	918758
919474	919800
922161	922537
926405	926764
926970	927384
928795	929152
933671	933792
935041	935298
935401	935810
936587	936774
938073	938330
957035	957401
958167	958622
968692	968924
971143	971395
975200	975602
976538	976865
978628	978669
981342	981861
982813	983186
990027	990372
990446	991296
994161	994697
995665	996401
999428	999742
1002572	1003057
1007153	1007573
1012039	1012541
1014828	1015224
1015534	1015842
1023192	1023631
1028457	1028814
1029271	1029585
1031131	1031548
1032442	1032783
1034070	1034513

Table S2. Putative promoter regions identified in *F. tularensis* LVS, continued

1034832	1034952
1036960	1037222
1045152	1045454
1046623	1047074
1049400	1049788
1054980	1055371
1055759	1056092
1056653	1056998
1058976	1059386
1064669	1065178
1066409	1066601
1073244	1073691
1073747	1074132
1075899	1076239
1078332	1078670
1081035	1081538
1083899	1084369
1085395	1086307
1096866	1097645
1102089	1102208
1102854	1103366
1107338	1107724
1118773	1119375
1125184	1125444
1130160	1130644
1130840	1131203
1135687	1136148
1138029	1138511
1138800	1139153
1140867	1141223
1144698	1145042
1148903	1149340
1152139	1152527
1155205	1155491
1162323	1162648
1166108	1166450
1167334	1168114
1168766	1170094
1170773	1171369
1172400	1172930
1173482	1173861
1174863	1175255
1179631	1180209
1182617	1182950
1183721	1184245
1185101	1185267
1189488	1189904
1194207	1194480
1198064	1198394
1198795	1199127
1199431	1199894
1200234	1200703
1201018	1201308
1202678	1203114
1206991	1207869
1208171	1208962
1210976	1211324
1212649	1213008
1219322	1219732
1226397	1226803
1229136	1229761
1232848	1233156
1235702	1236011
1237036	1237154
1238775	1239054
1240353	1240745
1242814	1243133
1245142	1245462
1248121	1248506
1249889	1250441
1252764	1253135
1253488	1253573
1254538	1256793
1265390	1265822
1269892	1270303
1270936	1271231
1271525	1271832
1273220	1273522
1281387	1282436
1285349	1285695
1287449	1287793
1290076	1290298

Table S2. Putative promoter regions identified in *F. tularensis* LVS, continued

1292837	1293337
1294192	1294878
1297799	1298395
1305819	1306005
1307782	1308075
1312938	1313088
1313824	1314141
1321406	1321705
1323043	1323402
1323447	1323976
1326399	1326789
1331579	1332087
1333737	1334208
1335637	1336061
1338573	1338966
1339710	1340164
1342912	1343236
1347806	1348196
1349815	1350158
1355709	1356691
1368356	1368639
1370133	1370556
1378393	1378758
1379363	1379901
1384098	1384506
1387594	1387821
1390452	1390828
1391476	1391736
1394364	1394740
1395390	1395651
1402493	1402813
1404512	1404894
1406043	1406550
1411111	1411286
1411628	1411982
1412060	1412486
1412782	1413143
1414124	1414635
1417328	1417773
1420436	1420738
1421424	1421763
1421850	1422188
1423764	1424081
1430682	1431466
1434136	1434417
1436467	1437010
1443043	1443449
1443859	1444323
1446225	1446510
1449062	1449374
1451604	1452178
1457976	1458292
1459357	1460299
1462920	1463789
1464869	1465250
1467571	1468143
1472830	1473356
1473777	1474051
1476168	1476556
1478634	1479151
1481471	1481844
1484318	1484741
1486626	1486989
1487764	1488377
1490249	1490591
1500159	1500434
1506551	1506967
1509797	1510364
1512053	1512371
1517746	1518060
1518765	1519216
1522209	1522559
1526892	1527347
1529171	1529880
1532928	1533334
1535095	1535560
1536919	1537185
1539958	1540346
1542391	1542748
1544462	1544756
1546046	1546513
1549865	1550230

Table S2. Putative promoter regions identified in *F. tularensis* LVS, continued

1553100	1553837
1554233	1554541
1556429	1556726
1560941	1561204
1569242	1569625
1570063	1570689
1572117	1572402
1573301	1573388
1575674	1575956
1577625	1578032
1584290	1584577
1586668	1586967
1589041	1589421
1599299	1599655
1601662	1602046
1602152	1602567
1603245	1603719
1603941	1604284
1608508	1608875
1611369	1611997
1612560	1613651
1614521	1614755
1622482	1622866
1623852	1624198
1624641	1625167
1628284	1628691
1631588	1631910
1635178	1635524
1635857	1636296
1639278	1639591
1641632	1642160
1642482	1643117
1647342	1647676
1648024	1648806
1653858	1654693
1657243	1657652
1659817	1660214
1660973	1661598
1665148	1665428
1667084	1667473
1668638	1669145
1671371	1671919
1682451	1682941
1683712	1684012
1684739	1685104
1686469	1687062
1689795	1690040
1691443	1691867
1695540	1696000
1700522	1701296
1706038	1706359
1709316	1710125
1710643	1711007
1720794	1721336
1724525	1724985
1725543	1725947
1727117	1727568
1727863	1728239
1734628	1735475
1739079	1739281
1746068	1746856
1764452	1764983
1772671	1773226
1777784	1778125
1786167	1786521
1797680	1798255
1800491	1801293
1803374	1803648
1804150	1804378
1806653	1807061
1818563	1818668
1824004	1826119
1826211	1826578
1828346	1828725
1830615	1831153
1832110	1832523
1833076	1833444
1835800	1836255
1842214	1842652
1844082	1844780
1846748	1847288
1848730	1849097

Table S2. Putative promoter regions identified in *F. tularensis* LVS, continued

1849201	1849655
1850896	1851206
1851374	1851714
1852021	1852629
1856664	1857099
1861401	1861878
1867980	1868321
1876656	1877042
1878519	1879366
1879914	1882737
1882924	1895368

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	17.14	1347866	1347260	1348175	915	FTL_1417	518	-	-	major facilitator transporter
1	17.14	1347866	1347260	1348175	915	FTL_R0035	268	-	*tRNA-Leu4*	Leu tRNA
1	17.14	1347866	1347260	1348175	915	FTL_R0036	186	-	*tRNA-His1*	His tRNA
1	17.14	1347866	1347260	1348175	915	FTL_R0037	82	-	*tRNA-Arg2*	Arg tRNA
1	17.14	1347866	1347260	1348175	915	FTL_R0038	-3	-	*tRNA-Pro1*	Pro tRNA
1	17.14	1347866	1347260	1348175	915	FTL_R0039	-96	-	*tRNA-Phe1*	Phe tRNA
2	15.07	643983	643678	644340	662	FTL_R0025	-22	+	*tRNA-Cys1*	Cys tRNA
2	15.07	643983	643678	644340	662	FTL_R0026	57	+	*tRNA-Leu2*	Leu tRNA
3	14.55	1894121	1893900	1894451	551	FTL_1966	604	-	-	anthranilate synthase component I
3	14.55	1894121	1893900	1894451	551	FTL_1967	308	-	-	trp operon repressor
3	14.55	1894121	1893900	1894451	551	FTL_R0051	-24	-	*tRNA-Asp1*	Asp tRNA
4	11.86	223331	222966	223647	681	FTL_R0009	-9	+	*tRNA-Glu1*	Glu tRNA
5	11.72	415740	415300	420853	5553	FTL_0445	533	-	-	hypothetical protein
5	11.72	415740	415300	420853	5553	FTL_R0016	-44	+	-	16S ribosomal RNA
6	11.51	1119478	1118779	1124617	5838	FTL_1172	539	-	-	hypothetical protein
7	11.09	636551	636271	636856	585	FTL_0652	412	-	-	hypothetical protein
7	11.09	636551	636271	636856	585	FTL_0653	295	+	-	hypothetical protein
7	11.09	636551	636271	636856	585	FTL_0654	712	+	-	uroporphyrinogen III synthase
7	11.09	636551	636271	636856	585	FTL_R0023	-68	-	*tRNA-Asn1*	Asn tRNA
8	10.78	124952	124136	129717	5581	FTL_0126	652	-	-	hypothetical protein
9	9.31	293277	292796	294221	1425	FTL_0306	179	-	-	tryptophanyl-tRNA synthetase
9	9.31	293277	292796	294221	1425	FTL_0308	930	+	-	hypothetical protein
9	9.31	293277	292796	294221	1425	FTL_R0011	5	-	*tRNA-Gly3*	Gly tRNA

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

9	9.31	293277	292796	294221	1425	FTL_R0012	-86	-	*tRNA-Arg3*	Arg tRNA
10	8.86	1293137	1292891	1293355	464	FTL_1358	842	-	-	cation-efflux family protein
10	8.86	1293137	1292891	1293355	464	FTL_1359	182	-	-	hypothetical protein
10	8.86	1293137	1292891	1293355	464	FTL_R0034	-38	+	*tRNA-Ser3*	Ser tRNA
11	8.28	67551	66769	67911	1142	FTL_0070	31	-	*rpsT*	30S ribosomal protein S20
11	8.28	67551	66769	67911	1142	FTL_0071	239	+	-	GTP-binding protein LepA
12	8.15	1414390	1414104	1414630	526	FTL_R0040	-40	-	*tRNA-Lys1*	Lys tRNA
13	7.64	414322	413879	414660	781	FTL_0444	221	-	*metG*	methionyl-tRNA synthetase
13	7.64	414322	413879	414660	781	FTL_R0014	-42	+	*tRNA-Ser2*	Ser tRNA
13	7.64	414322	413879	414660	781	FTL_R0015	77	+	*tRNA-Val1*	Val tRNA
14	7.35	1746255	1745762	1746493	731	FTL_1810	614	-	*nusA*	transcription elongation factor NusA
14	7.35	1746255	1745762	1746493	731	FTL_1811	144	-	-	hypothetical protein
14	7.35	1746255	1745762	1746493	731	FTL_R0050	-8	-	*tRNA-Met2*	Met tRNA
15	7.33	1265516	1264086	1265845	1759	FTL_1328	-14	-	-	outer membrane associated protein
16	6.96	1642909	1641259	1643171	1912	FTL_1709	967	-	-	hypothetical protein
16	6.96	1642909	1641259	1643171	1912	FTL_1710	186	-	-	ProP osmoprotectant transporter, fragment
16	6.96	1642909	1641259	1643171	1912	FTL_R0042	-9	+	*tRNA-Met1*	Met tRNA
17	6.86	178202	177982	179437	1455	FTL_0173	605	-	-	shikimate 5-dehydrogenase
17	6.86	178202	177982	179437	1455	FTL_0175	72	+	*rpmH*	50S ribosomal protein L34
17	6.86	178202	177982	179437	1455	FTL_0176	196	+	-	ribonuclease P protein component
17	6.86	178202	177982	179437	1455	FTL_0177	525	+	-	hypothetical protein
17	6.86	178202	177982	179437	1455	FTL_0178	782	+	-	inner-membrane protein
18	6.75	1333858	1332405	1334283	1878	FTL_1404	827	-	*rplT*	50S ribosomal protein L20
18	6.75	1333858	1332405	1334283	1878	FTL_1405	594	-	*rpmI*	50S ribosomal protein L35
18	6.75	1333858	1332405	1334283	1878	FTL_1406	84	-	-	translation initiation factor IF-3
20	6.51	1463239	1462989	1463500	511	FTL_1531	843	-	*rumA*	23S rRNA 5-methyluridine

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

										methyltransferase
20	6.51	1463239	1462989	1463500	511	FTL_1532	182	-	-	hypothetical protein
20	6.51	1463239	1462989	1463500	511	FTL_R0041	-18	-	*tRNA-Val3*	Val tRNA
21	6.03	526964	526524	527219	695	FTL_R0021	-52	-	*tRNA-Met3*	Met tRNA
22	5.84	633220	632890	633464	574	FTL_0650	186	-	-	prolyl-tRNA synthetase
23	5.56	1667163	1666019	1667461	1442	FTL_1734	915	-	-	integrase/recombinase
23	5.56	1667163	1666019	1667461	1442	FTL_1735	402	-	*rplS*	50S ribosomal protein L19
24	5.51	389737	388570	391273	2703	FTL_0423	190	+	-	hypothetical protein
24	5.51	389737	388570	391273	2703	FTL_0424	726	+	-	lipoprotein
25	5.4	158547	158290	158872	582	FTL_0152	145	+	-	hypothetical protein
25	5.4	158547	158290	158872	582	FTL_0153	489	+	*isftu1*	transposase
25	5.4	158547	158290	158872	582	FTL_R0008	-25	+	*tRNA-Gln1*	Gln tRNA
26	5.13	1379500	1378637	1379804	1167	FTL_1452	365	-	*rpmA*	50S ribosomal protein L27
26	5.13	1379500	1378637	1379804	1167	FTL_1453	20	-	*rplU*	50S ribosomal protein L21
26	5.13	1379500	1378637	1379804	1167	FTL_1454	251	+	-	50S ribosomal protein L21, pseudogene
26	5.13	1379500	1378637	1379804	1167	FTL_1455	593	+	-	50S ribosomal protein L21, pseudogene
27	5.11	501746	499231	502033	2802	FTL_0520	438	-	-	septum site-determining protein MinC
27	5.11	501746	499231	502033	2802	FTL_0521	248	-	*rpmG*	50S ribosomal protein L33
27	5.11	501746	499231	502033	2802	FTL_0522	-15	-	*rpmB*	50S ribosomal protein L28
27	5.11	501746	499231	502033	2802	FTL_0524	623	+	-	ATP-dependent DNA helicase RecG
28	5.08	904813	904592	905009	417	FTL_0933	25	+	-	DNA recombination protein RmuC family protein
29	5.03	450239	449994	450514	520	FTL_0474	542	+	-	lipoprotein releasing system, subunit C,putative membrane protein
29	5.03	450239	449994	450514	520	FTL_R0053	-60	+	-	5S ribosomal RNA
30	5.01	75740	75360	75870	510	FTL_0078	136	-	-	riboflavin biosynthesis protein

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

										ribD
31	4.88	236518	230665	247823	17158	FTL_0240	339	+	*rpsS*	30S ribosomal protein S19
31	4.88	236518	230665	247823	17158	FTL_0241	633	+	*rplV*	50S ribosomal protein L22
31	4.88	236518	230665	247823	17158	FTL_0242	983	+	*rpsC*	30S ribosomal protein S3
32	4.88	1710558	1709985	1711007	1022	FTL_1777	997	-	-	hypothetical protein, pseudogene
32	4.88	1710558	1709985	1711007	1022	FTL_1778	543	-	-	hypothetical protein, pseudogene
32	4.88	1710558	1709985	1711007	1022	FTL_R0049	245	-	*tRNA-Leu3*	Leu tRNA
33	4.87	32007	31775	32194	419	FTL_0030	118	-	-	carbamoyl phosphate synthase small subunit
33	4.87	32007	31775	32194	419	FTL_0031	299	+	-	acid phosphatase
34	4.86	628167	627862	628819	957	FTL_0644	235	-	-	carbon-nitrogen hydrolase
34	4.86	628167	627862	628819	957	FTL_0645	-66	+	-	lipoprotein
35	4.75	1800645	1800397	1801032	635	FTL_1868	-20	-	-	multidrug resistance protein, membrane located
35	4.75	1800645	1800397	1801032	635	FTL_1869	195	+	-	Na+/H+ antiporter
36	4.53	1002991	1002442	1003914	1472	FTL_1046	289	-	-	D-alanyl-D-alanine carboxypeptidase
36	4.53	1002991	1002442	1003914	1472	FTL_1047	-33	+	*rpsU*	30S ribosomal protein S21
36	4.53	1002991	1002442	1003914	1472	FTL_1048	225	+	-	hypothetical protein
36	4.53	1002991	1002442	1003914	1472	FTL_1049	692	+	-	DNA primase
38	4.36	1526969	1526453	1527387	934	FTL_1597	-92	-	-	organic solvent tolerance protein
38	4.36	1526969	1526453	1527387	934	FTL_1598	218	+	*argS*	arginyl-tRNA synthetase
39	4.25	353740	352014	354164	2150	FTL_0383	313	+	-	mannose-6-phosphate isomerase, fragment
39	4.25	353740	352014	354164	2150	FTL_0384	768	+	-	mannose-6-phosphate isomerase, fragment, pseudogene
40	4.23	23980	22633	24709	2076	FTL_R0001	834	-	*ssrA*	-
41	4.11	1229592	1229438	1229799	361	FTL_1287	778	-	*rlmL*	23S rRNA m(2)G2445

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

										methyltransferase
41	4.11	1229592	1229438	1229799	361	FTL_1288	149	-	-	fatty acid hydroxylase
41	4.11	1229592	1229438	1229799	361	FTL_R0033	-17	+	*tRNA-Val2*	Val tRNA
42	4.05	786408	786064	787254	1190	FTL_0801	-41	+	*aroK*	shikimate kinase I
42	4.05	786408	786064	787254	1190	FTL_0802	486	+	*aroB*	3-dehydroquinate synthase
43	3.96	1135811	1133892	1136111	2219	FTL_1185	956	-	-	macrophage growth locus subunit A
43	3.96	1135811	1133892	1136111	2219	FTL_1186	459	-	*rpsI*	30S ribosomal protein S9
43	3.96	1135811	1133892	1136111	2219	FTL_1187	15	-	*rplM*	50S ribosomal protein L13
44	3.94	1648476	1647988	1648776	788	FTL_1717	174	+	-	hypothetical protein
44	3.94	1648476	1647988	1648776	788	FTL_R0044	-93	-	*ffs*	-
45	3.94	362105	361818	362530	712	FTL_0391	902	-	-	hypothetical protein
45	3.94	362105	361818	362530	712	FTL_0393	592	+	-	hypothetical protein
46	3.88	1467793	1466440	1468119	1679	FTL_1537	289	-	-	polynucleotide phosphorylase/polyadenylase
46	3.88	1467793	1466440	1468119	1679	FTL_1538	-34	-	*rpsO*	30S ribosomal protein S15
47	3.85	607107	605835	608434	2599	FTL_R0022	-71	+	*tRNA-Leu1*	Leu tRNA
48	3.81	84090	83040	84314	1274	FTL_0088	501	-	-	acetyltransferase protein
48	3.81	84090	83040	84314	1274	FTL_0089	15	-	-	ferredoxin
48	3.81	84090	83040	84314	1274	FTL_0090	218	+	-	hypothetical protein
50	3.78	680778	680522	681051	529	FTL_0691	-40	-	-	proton-dependent oligopeptide transport (POT) family protein
51	3.66	1668830	1667975	1669109	1134	FTL_1737	399	-	*rimM*	16S rRNA-processing protein RimM
51	3.66	1668830	1667975	1669109	1134	FTL_1738	108	-	*rpsP*	30S ribosomal protein S16
51	3.66	1668830	1667975	1669109	1134	FTL_1739	186	+	-	S-adenosylmethionine synthetase
52	3.66	1240701	1240358	1241163	805	FTL_1303	-47	+	*rpmE*	50S ribosomal protein L31
53	3.59	983313	982835	984411	1576	FTL_1025	96	+	*rpsR*	30S ribosomal protein S18
53	3.59	983313	982835	984411	1576	FTL_1026	333	+	*rplI*	50S ribosomal protein L9

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

53	3.59	983313	982835	984411	1576	FTL_1027	879	+	-	replicative DNA helicase
54	3.48	1051610	1051343	1051999	656	FTL_1107	712	+	-	hypothetical protein
55	3.43	142733	142471	142987	516	FTL_0136	290	-	-	hypothetical protein
57	3.36	1731872	1728144	1735613	7469	FTL_1796	932	-	-	F0F1 ATP synthase subunit gamma
59	3.31	314833	314311	315145	834	FTL_0330	174	-	-	hypothetical protein
59	3.31	314833	314311	315145	834	FTL_0331	-4	+	-	TolQ protein
59	3.31	314833	314311	315145	834	FTL_0332	719	+	-	TolR protein
60	3.3	828185	827965	828985	1020	FTL_0847	475	+	-	preprotein translocase family protein
60	3.3	828185	827965	828985	1020	FTL_0848	888	+	*secD*	preprotein translocase subunit SecD
61	3.29	1781136	1780200	1781432	1232	FTL_1847	846	-	-	toxin secretion ABC transporter ATP-binding protein
61	3.29	1781136	1780200	1781432	1232	FTL_1848	521	-	-	hypothetical protein
62	3.23	1589212	1588918	1589434	516	FTL_1658	4	-	-	hypothetical protein
63	3.2	534392	534035	535251	1216	FTL_0552	-76	+	-	two-component response regulator
63	3.2	534392	534035	535251	1216	FTL_0553	615	+	-	signal peptidase I
64	3.18	769599	769475	769892	417	FTL_0783	225	-	*isftu1*	transposase
65	3.16	8235	7977	9286	1309	FTL_0009	-41	+	-	outer membrane protein
65	3.16	8235	7977	9286	1309	FTL_0010	634	+	-	thiosulfate sulfurtransferase
66	3.12	1602277	1601683	1602547	864	FTL_1668	-46	-	-	lipid A transport protein ABC transporter ATP-binding protein/permease
66	3.12	1602277	1601683	1602547	864	FTL_1669	151	+	-	tRNA CCA-pyrophosphorylase
67	3.11	662489	660408	662694	2286	FTL_0674	606	-	*panB*	3-methyl-2-oxobutanoate hydroxymethyltransferase
67	3.11	662489	660408	662694	2286	FTL_0675	-55	-	-	hypothetical protein
67	3.11	662489	660408	662694	2286	FTL_0676	363	+	-	DNA ligase
68	3.1	919286	918435	920273	1838	FTL_0948	956	-	-	hypothetical protein

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

68	3.1	919286	918435	920273	1838	FTL_0950	403	+	-	50S ribosomal protein L25
68	3.1	919286	918435	920273	1838	FTL_0951	789	+	-	hypothetical protein
69	3.08	133987	133684	134310	626	FTL_0131	195	-	-	branched-chain amino acid aminotransferase
70	3.08	1014955	1014792	1015205	413	FTL_1060	5	-	-	D-alanyl-D-alanine carboxypeptidase
70	3.08	1014955	1014792	1015205	413	FTL_1061	152	+	-	inorganic pyrophosphatase
70	3.08	1014955	1014792	1015205	413	FTL_1062	764	+	-	3-deoxy-D-manno-octulosonate 8-phosphate phosphatase
71	3.06	221706	221536	221999	463	FTL_0220	475	-	*isftu2*	transposase
71	3.06	221706	221536	221999	463	FTL_0221	-32	+	-	amino acid permease
72	3.05	1152401	1152268	1152542	274	FTL_1202	198	-	-	hypothetical protein
73	2.99	136807	136623	136980	357	FTL_0132	-19	-	-	pyruvate phosphate dikinase
74	2.97	521543	521207	523454	2247	FTL_0538	190	+	*fabZ*	(3R)-hydroxymyristoyl-ACP dehydratase
74	2.97	521543	521207	523454	2247	FTL_0539	684	+	-	UDP-N-acetylglucosamine acyltransferase
75	2.97	1080453	1079302	1081473	2171	FTL_1136	900	-	-	hypothetical protein
76	2.97	1208513	1208261	1209102	841	FTL_1266	376	+	-	lipase/esterase
78	2.94	56989	56756	57176	420	FTL_0056	190	-	-	NADH dehydrogenase
78	2.94	56989	56756	57176	420	FTL_0057	213	+	-	hypothetical protein
79	2.89	709724	709534	709992	458	FTL_0717	121	-	-	ribonuclease E
79	2.89	709724	709534	709992	458	FTL_0718	129	+	-	cysteine desulfarase
80	2.88	1183998	1183504	1184238	734	FTL_1236	-1	-	*infA*	translation initiation factor IF-1
80	2.88	1183998	1183504	1184238	734	FTL_1237	161	+	-	hypothetical protein
80	2.88	1183998	1183504	1184238	734	FTL_1238	997	+	-	hypothetical protein
82	2.85	1478775	1478502	1479107	605	FTL_1547	-36	-	-	DNA gyrase subunit B
82	2.85	1478775	1478502	1479107	605	FTL_1548	218	+	-	hypothetical protein
83	2.84	181679	181399	182004	605	FTL_0180	-73	+	-	acyltransferase

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

83	2.84	181679	181399	182004	605	FTL_0181	863	+	-	Type IV pili fiber building block protein
84	2.84	1654026	1653755	1654529	774	FTL_1722	-8	-	-	glutamyl-tRNA reductase
84	2.84	1654026	1653755	1654529	774	FTL_1723	188	+	-	hypothetical protein
84	2.84	1654026	1653755	1654529	774	FTL_1724	829	+	-	lipoprotein
85	2.84	1762368	1760724	1764782	4058	FTL_1826	526	-	-	NADH dehydrogenase I subunit E
86	2.8	886434	886253	886651	398	FTL_0913	84	+	-	ribosome biogenesis GTP-binding protein YsxC, pseudogene
87	2.79	476984	476767	477372	605	FTL_0491	-22	+	-	outer membrane lipoprotein
87	2.79	476984	476767	477372	605	FTL_0492	726	+	-	UDP-N--acetylmuramoylalanyl-D-glutamyl-2,6- diaminopimelate-D-alanyl-D-alanyl ligase
88	2.76	602509	601979	602871	892	FTL_0610	352	-	*rho*	transcription termination factor Rho
88	2.76	602509	601979	602871	892	FTL_0611	-14	-	-	thioredoxin
88	2.76	602509	601979	602871	892	FTL_0612	212	+	-	exopolyphosphatase
89	2.75	1842336	1841855	1842630	775	FTL_1908	-12	-	-	cell division protein FtsA
90	2.74	1305678	1304990	1305940	950	FTL_1372	-5	-	-	lipoprotein
90	2.74	1305678	1304990	1305940	950	FTL_1373	478	+	-	hypothetical protein
91	2.72	996062	995920	996420	500	FTL_1039	351	+	-	hypothetical protein
91	2.72	996062	995920	996420	500	FTL_R0027	192	+	-	-
92	2.72	278303	278015	278838	823	FTL_0292	375	-	-	GTP pyrophosphokinase, pseudogene
93	2.71	839534	839220	839785	565	FTL_0858	79	-	-	hypothetical protein
94	2.7	268144	267858	268288	430	FTL_0282	269	-	-	hypothetical protein
94	2.7	268144	267858	268288	430	FTL_0283	-78	+	-	aromatic amino acid HAAP transporter
95	2.69	1630150	1629936	1630376	440	FTL_1699	65	-	-	hypothetical protein
96	2.68	1671504	1670371	1671809	1438	FTL_1740	-50	-	-	fatty acid desaturase

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

97	2.66	1170729	1168684	1171105	2421	FTL_1220	570	+	-	amino-acid permease
98	2.66	19349	18621	19616	995	FTL_0020	-32	-	*aspS*	aspartyl-tRNA synthetase
99	2.64	874196	873872	874564	692	FTL_0896	272	+	-	hypothetical protein
100	2.63	513919	513688	514187	499	FTL_0531	864	-	-	hypothetical protein
100	2.63	513919	513688	514187	499	FTL_0532	284	-	-	hypothetical protein
100	2.63	513919	513688	514187	499	FTL_0533	-25	+	-	DNA gyrase subunit A
101	2.61	1553354	1552996	1553531	535	FTL_1622	164	-	-	multidrug transporter (tetracycline resistance protein)
102	2.58	1404605	1404373	1404915	542	FTL_1478	-41	-	-	inosine-5-monophosphate dehydrogenase
103	2.56	843987	843766	844250	484	FTL_0863	-20	-	-	Sodium-dicarboxylate symporter family protein
103	2.56	843987	843766	844250	484	FTL_0864	222	+	-	SIS domain-containing protein
104	2.56	729634	729410	729792	382	FTL_0740	267	+	-	hypothetical protein
105	2.56	1825656	1825246	1826052	806	FTL_1892	639	-	-	hypothetical protein
106	2.55	425091	424911	425651	740	FTL_0449	-66	+	-	hypothetical protein
107	2.55	1724040	1722342	1724272	1930	FTL_1792	768	+	-	glutaredoxin-like protein
108	2.53	530961	530646	531199	553	FTL_0547	17	-	-	3-deoxy-D-manno-octulosonic-acid transferase
108	2.53	530961	530646	531199	553	FTL_0549	773	+	-	pyrroline-5-carboxylate reductase
110	2.5	1356233	1356087	1356486	399	FTL_1425	275	-	-	hypothetical protein
111	2.5	877283	876777	877560	783	FTL_0899	34	+	-	protease, GTP-binding subunit
112	2.48	225762	224858	226450	1592	FTL_0223	755	-	-	dihydrofolate reductase type I
112	2.48	225762	224858	226450	1592	FTL_0225	179	+	*tsf*	elongation factor Ts
114	2.44	1431058	1430697	1431454	757	FTL_1499	319	-	-	sulfate permease family protein
114	2.44	1431058	1430697	1431454	757	FTL_1502	650	+	-	major facilitator transporter
115	2.42	1465043	1464793	1465500	707	FTL_1535	7	-	-	2-dehydro-3-deoxyphosphooctonate aldolase

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

118	2.41	1297986	1297497	1298393	896	FTL_1364	-17	-	-	hypothetical protein
118	2.41	1297986	1297497	1298393	896	FTL_1365	418	+	-	hypothetical protein
118	2.41	1297986	1297497	1298393	896	FTL_1366	942	+	-	membrane fusion protein
119	2.39	1213270	1212613	1214230	1617	FTL_1271	497	-	-	adenosylmethionine-8-amino-7-oxononanoate aminotransferase
119	2.39	1213270	1212613	1214230	1617	FTL_1273	529	+	-	8-amino-7-oxononanoate synthase
120	2.37	1806765	1806626	1807000	374	FTL_1873	984	-	-	amino acid transporter
120	2.37	1806765	1806626	1807000	374	FTL_1874	-27	-	*obgE*	GTPase ObgE
121	2.37	1647296	1646790	1647591	801	FTL_1714	259	-	*groEL*	chaperonin GroEL
121	2.37	1647296	1646790	1647591	801	FTL_1715	-66	-	*groES*	co-chaperonin GroES
122	2.36	1796059	1795999	1796113	114	FTL_1864	433	-	-	hypothetical protein
123	2.36	256389	256031	256554	523	FTL_0269	49	-	-	glutamate dehydrogenase
123	2.36	256389	256031	256554	523	FTL_0270	437	+	*isftu2*	transposase
124	2.35	1797803	1796364	1798236	1872	FTL_1865	639	-	-	outer membrane protein tolC
124	2.35	1797803	1796364	1798236	1872	FTL_1866	10	-	-	protein-L-isoaspartate O-methyltransferase
124	2.35	1797803	1796364	1798236	1872	FTL_1867	310	+	-	protease yegQ
125	2.35	660043	659513	660383	870	FTL_0671	116	-	-	pantothenate kinase
126	2.34	432342	431901	432905	1004	FTL_0456	-12	-	*rpsU*	30S ribosomal protein S21
126	2.34	432342	431901	432905	1004	FTL_0459	918	+	-	methionine aminopeptidase
127	2.32	781747	781420	781978	558	FTL_0795	173	-	*adk*	adenylate kinase
127	2.32	781747	781420	781978	558	FTL_0796	-46	+	-	hypothetical protein
127	2.32	781747	781420	781978	558	FTL_0797	960	+	-	Type IV pili associated protein
128	2.3	1012153	1012065	1012424	359	FTL_1056	892	-	-	DNA primase, pseudogene
128	2.3	1012153	1012065	1012424	359	FTL_1057	-8	-	-	DNA primase, pseudogene
129	2.3	319277	319012	319572	560	FTL_0337	-64	+	-	peptidoglycan-associated lipoprotein, pseudogene
130	2.28	153045	152812	153253	441	FTL_0147	6	+	-	hypothetical protein

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

131	2.27	1249987	1249839	1250089	250	FTL_1311	-23	-	*pyrG*	CTP synthetase
131	2.27	1249987	1249839	1250089	250	FTL_1312	336	+	-	S-transferase
133	2.25	1097561	1097332	1097809	477	FTL_1154	858	-	-	transketolase, pseudogene
133	2.25	1097561	1097332	1097809	477	FTL_1157	-70	+	-	intracellular growth locus, subunit A
133	2.25	1097561	1097332	1097809	477	FTL_1158	498	+	-	intracellular growth locus, subunit B
134	2.25	162575	162310	162996	686	FTL_0156	822	-	-	phosphate transport protein
134	2.25	162575	162310	162996	686	FTL_0157	148	-	-	hypothetical protein
134	2.25	162575	162310	162996	686	FTL_0158	159	+	-	acid phosphatase
135	2.24	964263	964015	964522	507	FTL_0994	788	-	-	hypothetical protein
135	2.24	964263	964015	964522	507	FTL_0995	152	-	-	haloacid dehalogenase
135	2.24	964263	964015	964522	507	FTL_0996	-56	+	-	AhpC/Tsa family protein
135	2.24	964263	964015	964522	507	FTL_0997	608	+	-	hypothetical protein
136	2.24	1185435	1185286	1185717	431	FTL_1239	-93	+	-	signal recognition particle protein, Ffh
139	2.23	1207635	1207293	1208005	712	FTL_1263	461	-	-	AMP-binding family protein
140	2.23	1085257	1083007	1086292	3285	FTL_1142	64	-	-	putative glycerol-3-phosphate acyltransferase PlsX
140	2.23	1085257	1083007	1086292	3285	FTL_1145	903	+	-	transketolase
141	2.22	1661056	1660453	1661551	1098	FTL_1729	-72	-	-	dinucleoside polyphosphate hydrolase
141	2.22	1661056	1660453	1661551	1098	FTL_1730	511	+	-	hypothetical protein
143	2.2	1628460	1628302	1628728	426	FTL_1697	817	-	-	metal ion transporter
143	2.2	1628460	1628302	1628728	426	FTL_1698	243	-	-	hypothetical protein
144	2.19	269905	269268	270119	851	FTL_0285	116	+	-	GTP pyrophosphokinase
146	2.18	701813	701700	701933	233	FTL_0710	512	-	-	choloylglycine hydrolase family protein
146	2.18	701813	701700	701933	233	FTL_0711	-16	+	-	proton-dependent oligopeptide transport (POT) family protein
146	2.18	701813	701700	701933	233	FTL_0712	668	+	-	hypothetical protein

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

147	2.15	824417	824195	824510	315	FTL_0842	168	-	-	transposase
147	2.15	824417	824195	824510	315	FTL_0843	138	+	*tgt*	queuine tRNA-ribosyltransferase
148	2.15	1459877	1459669	1460073	404	FTL_1528	455	-	-	major facilitator transporter
149	2.14	249643	248613	250568	1955	FTL_0264	430	-	-	30S ribosomal protein S3, pseudogene
149	2.14	249643	248613	250568	1955	FTL_0265	-73	+	-	hypothetical protein
151	2.13	935702	935460	935817	357	FTL_0967	146	-	-	lipoate-protein ligase A
151	2.13	935702	935460	935817	357	FTL_0968	-32	+	-	tyrosyl-tRNA synthetase
152	2.13	98963	97829	99727	1898	FTL_0102	747	-	-	hypothetical protein
152	2.13	98963	97829	99727	1898	FTL_0103	279	-	-	hypothetical protein
152	2.13	98963	97829	99727	1898	FTL_0106	770	+	-	hypothetical protein, pseudogene
153	2.1	1635039	1634702	1635470	768	FTL_1703	-81	-	-	amino acid transporter
154	2.1	838687	838358	838835	477	FTL_0857	168	-	-	hypothetical protein
155	2.1	1142162	1142022	1142393	371	FTL_1194	920	+	-	pirin family protein
156	2.09	1023568	1023388	1023740	352	FTL_1071	-89	+	*guaA*	GMP synthase
157	2.08	176670	176539	176841	302	FTL_0172	-87	-	-	UDP-N-acetylmuramate--L-alanine ligase
158	2.08	1499299	1499234	1499383	149	FTL_1571	-80	+	-	thioredoxin reductase
159	2.08	1166189	1166012	1166423	411	FTL_1215	555	-	-	hypothetical protein
159	2.08	1166189	1166012	1166423	411	FTL_1216	-31	-	-	hypothetical protein
160	2.07	714445	714136	714564	428	FTL_0723	735	-	-	hypothetical protein
160	2.07	714445	714136	714564	428	FTL_0724	184	-	-	5-formyltetrahydrofolate cyclo-ligase
160	2.07	714445	714136	714564	428	FTL_0725	256	+	-	hypothetical protein
160	2.07	714445	714136	714564	428	FTL_0726	801	+	-	2-octaprenyl-6-methoxyphenyl hydroxylase
161	2.06	1273218	1272978	1273382	404	FTL_1337	-63	-	-	phosphopantetheine adenylyltransferase, pseudogene

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

162	2.05	975458	975234	975627	393	FTL_1014	76	-	-	oxidative stress transcriptional regulator
162	2.05	975458	975234	975627	393	FTL_1015	22	+	-	AhpC/TSA family protein
162	2.05	975458	975234	975627	393	FTL_1016	549	+	-	short chain dehydrogenase
164	2.04	228383	228187	228504	317	FTL_0229	587	+	-	phosphatidate cytidylyltransferase
165	2.04	676487	676326	676749	423	FTL_0688	-71	-	-	major facilitator transporter
165	2.04	676487	676326	676749	423	FTL_0689	236	+	-	transcriptional regulator araC family protein
166	2.04	102944	102674	103179	505	FTL_0108	880	-	-	hypothetical protein
166	2.04	102944	102674	103179	505	FTL_0111	-92	+	-	intracellular growth locus, subunit A
166	2.04	102944	102674	103179	505	FTL_0112	476	+	-	intracellular growth locus, subunit B
167	2.03	495689	495527	496053	526	FTL_0510	817	-	*isftu1*	transposase
168	2.02	1486702	1486583	1486970	387	FTL_1555	132	+	-	hypothetical protein
168	2.02	1486702	1486583	1486970	387	FTL_1556	956	+	-	hypothetical protein
169	2.02	190350	189765	190553	788	FTL_0189	65	-	-	cytochrome d terminal oxidase, polypeptide subunit I
170	2	226750	226523	227118	595	FTL_0226	64	+	*pyrH*	uridylate kinase
171	1.99	865724	865404	865865	461	FTL_0886	222	-	-	(dimethylallyl)adenosine tRNA methylthiotransferase
171	1.99	865724	865404	865865	461	FTL_0887	1	+	-	o-methyltransferase family protein
171	1.99	865724	865404	865865	461	FTL_0888	866	+	-	o-methyltransferase family protein, pseudogene
172	1.99	1535345	1534896	1535553	657	FTL_1605	699	-	-	putative periplasmic protease
172	1.99	1535345	1534896	1535553	657	FTL_1606	58	-	-	stringent starvation protein A
172	1.99	1535345	1534896	1535553	657	FTL_1607	31	+	-	anaerobic sulfite reductase subunit
172	1.99	1535345	1534896	1535553	657	FTL_1608	366	+	-	hypothetical protein
172	1.99	1535345	1534896	1535553	657	FTL_1609	721	+	-	dolichyl-phosphate-mannose-protein mannosyltransferase

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

										family protein
173	1.98	282329	281424	282817	1393	FTL_0294	935	-	-	DNA mismatch repair protein MutS
173	1.98	282329	281424	282817	1393	FTL_0296	199	+	-	hypothetical protein
173	1.98	282329	281424	282817	1393	FTL_0297	683	+	-	hypothetical protein
174	1.98	1844419	1843865	1846517	2652	FTL_1910	399	-	-	D-alanyl-alanine synthetase A
174	1.98	1844419	1843865	1846517	2652	FTL_1911	-66	-	-	zinc-binding domain-containing protein
174	1.98	1844419	1843865	1846517	2652	FTL_1912	214	+	*rpsA*	30S ribosomal protein S1
175	1.97	555866	555784	556021	237	FTL_0578	468	+	-	ornithine cyclodeaminase
177	1.97	327852	327591	327947	356	FTL_0348	410	+	-	Ser tRNA, pseudogene
177	1.97	327852	327591	327947	356	FTL_R0013	-82	+	*tRNA-Ser1*	Ser tRNA
178	1.97	1458028	1457900	1458233	333	FTL_1527	-47	-	*eno*	phosphopyruvate hydratase
181	1.95	1866447	1865327	1866863	1536	FTL_1935	554	-	-	ABC transporter ATP-binding protein
182	1.95	894499	894343	894655	312	FTL_0918	715	-	-	hypothetical protein
183	1.95	1247870	1246700	1248383	1683	FTL_1308	954	-	-	bifunctional folylpolyglutamate synthase/ dihydrofolate synthase
183	1.95	1247870	1246700	1248383	1683	FTL_1309	37	-	-	Acetyl-CoA carboxylase beta subunit
185	1.9	372373	372208	372670	462	FTL_0401	730	-	-	Sua5_yciO_yrdC family protein
185	1.9	372373	372208	372670	462	FTL_0403	-65	+	-	hypothetical protein
186	1.9	584726	584473	585063	590	FTL_0598	51	+	-	membrane protein/O-antigen protein
187	1.9	1709498	1709358	1709700	342	FTL_1776	406	-	-	hypothetical protein
188	1.9	805505	805355	805761	406	FTL_0821	260	-	*hemH*	ferrochelatase
188	1.9	805505	805355	805761	406	FTL_0823	94	+	-	hypothetical protein
188	1.9	805505	805355	805761	406	FTL_0824	504	+	-	hypothetical protein, pseudogene
188	1.9	805505	805355	805761	406	FTL_0825	988	+	-	hypothetical protein,

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

										pseudogene
189	1.89	1484796	1484617	1484871	254	FTL_1552	309	-	-	hypothetical protein
190	1.89	1401006	1400550	1401214	664	FTL_1474	767	-	*greA*	transcription elongation factor GreA
190	1.89	1401006	1400550	1401214	664	FTL_1475	149	-	-	Type IV pili fiber building block protein
191	1.88	832129	831871	832642	771	FTL_0851	430	+	-	RNA polymerase factor sigma-32
192	1.88	580624	578806	584182	5376	FTL_0594	-35	+	-	UDP-glucose 4-epimerase
192	1.88	580624	578806	584182	5376	FTL_0595	746	+	-	galacturonosyl transferase
193	1.88	1546168	1545995	1546291	296	FTL_1616	-74	-	-	phosphoenolpyruvate carboxykinase
193	1.88	1546168	1545995	1546291	296	FTL_1617	225	+	-	glutaminyl-tRNA synthetase
194	1.86	470060	469973	470162	189	FTL_0487	444	+	-	maltodextrin phosphorylase
195	1.86	911903	911208	912173	965	FTL_0940	728	-	-	methylpurine-DNA glycosylase family protein
195	1.86	911903	911208	912173	965	FTL_0942	-97	+	-	nicotinamide mononucleotide transport (NMT) family protein
195	1.86	911903	911208	912173	965	FTL_0943	654	+	-	Sodium/hydrogen exchanger family protein
196	1.86	36443	36015	36922	907	FTL_0035	805	+	-	hypothetical protein
197	1.85	721348	720630	721829	1199	FTL_0732	368	+	-	lactoylglutathione lyase
198	1.83	1349899	1349770	1349994	224	FTL_1419	-18	-	-	cyanophycinase
198	1.83	1349899	1349770	1349994	224	FTL_1420	221	+	-	carbohydrate/purine kinase pfkB family protein
201	1.82	1441939	1441766	1442445	679	FTL_1510	-71	-	-	glycerol-3-phosphate transporter
202	1.82	1841027	1840664	1841848	1184	FTL_1907	-15	-	-	cell division protein FtsZ
203	1.82	1244873	1244016	1245416	1400	FTL_1305	693	-	-	virulence factor MviN
205	1.82	788706	788053	788791	738	FTL_0803	1	-	-	hypothetical protein
205	1.82	788706	788053	788791	738	FTL_0804	213	+	-	hypothetical protein
205	1.82	788706	788053	788791	738	FTL_0805	517	+	-	bifunctional proline dehydrogenase/pyrroline-5-

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

										carboxylate dehydrogenase
206	1.81	524321	523951	524844	893	FTL_0542	564	+	-	hypothetical protein
207	1.8	363582	363334	363887	553	FTL_0394	-26	+	-	bifunctional 5,10-methylene-tetrahydrofolate dehydrogenase/ 5,10-methylene-tetrahydrofolate cyclohydrolase
212	1.77	149690	149313	150058	745	FTL_0144	-11	-	-	hypothetical protein
212	1.77	149690	149313	150058	745	FTL_0145	140	+	-	ABC transporter membrane protein
213	1.77	1512378	1512081	1512461	380	FTL_1585	479	+	-	single-stranded-DNA-specific exonuclease
215	1.77	315237	315193	315285	92	FTL_0333	742	+	-	hypothetical protein
216	1.76	1189466	1189240	1189587	347	FTL_1241	536	-	*isftu1*	transposase
216	1.76	1189466	1189240	1189587	347	FTL_1242	279	-	-	ThiJ/Pfpl family protein
216	1.76	1189466	1189240	1189587	347	FTL_1243	-44	-	-	hypothetical protein
216	1.76	1189466	1189240	1189587	347	FTL_1244	328	+	-	exodeoxyribonuclease III
218	1.76	1179924	1179596	1180135	539	FTL_1230	528	-	-	cysteine desulfurase activator complex subunit SufB
218	1.76	1179924	1179596	1180135	539	FTL_1231	111	-	-	hypothetical protein
218	1.76	1179924	1179596	1180135	539	FTL_1232	42	+	*trkA*	potassium transporter peripheral membrane protein
219	1.75	1596809	1596543	1596929	386	FTL_1664	867	-	-	phosphopentomutase
219	1.75	1596809	1596543	1596929	386	FTL_1665	98	-	-	pantothenate kinase
220	1.75	207376	207218	207579	361	FTL_0207	-41	+	-	pyrrolidone-carboxylate peptidase
220	1.75	207376	207218	207579	361	FTL_0208	635	+	-	putative cytochrome c-type biogenesis protein
221	1.75	48009	47882	48239	357	FTL_0044	469	-	-	transglutaminase
221	1.75	48009	47882	48239	357	FTL_0045	-49	+	-	orotidine 5'-phosphate decarboxylase
221	1.75	48009	47882	48239	357	FTL_0046	585	+	-	diyroorotate dehydrogenase

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

222	1.75	1326462	1326415	1326790	375	FTL_1395	-81	-	-	major facilitator superfamily galactose-proton symporter
222	1.75	1326462	1326415	1326790	375	FTL_1396	172	+	-	galactose-1-phosphate uridylyltransferase
223	1.74	922166	921992	922286	294	FTL_0952	481	-	-	hypothetical protein
226	1.73	168800	168433	169571	1138	FTL_0164	498	-	*isftu1*	transposase
226	1.73	168800	168433	169571	1138	FTL_0165	254	-	-	dicarboxylate MFS transporter
227	1.73	1064824	1064677	1064990	313	FTL_1118	32	-	-	hypothetical protein
227	1.73	1064824	1064677	1064990	313	FTL_1119	162	+	*rdgC*	recombination associated protein
228	1.73	297439	297343	297514	171	FTL_0310	818	+	-	dihydrolipoamide acetyltransferase
229	1.72	1861800	1861685	1862100	415	FTL_1929	198	-	*purH*	bifunctional phosphoribosylaminoimidazole carboxamide formyltransferase/IMP cyclohydrolase
229	1.72	1861800	1861685	1862100	415	FTL_1930	-21	+	-	adenylosuccinate synthetase
230	1.72	1175438	1175332	1176195	863	FTL_1225	400	-	-	hypothetical protein
230	1.72	1175438	1175332	1176195	863	FTL_1226	-23	-	-	hypothetical protein, pseudogene
231	1.72	880286	880207	880401	194	FTL_0901	638	-	-	monooxygenase family protein
231	1.72	880286	880207	880401	194	FTL_0902	474	-	-	oxidoreductase
231	1.72	880286	880207	880401	194	FTL_0904	780	+	-	hypothetical protein
233	1.7	818918	818799	819042	243	FTL_0835	175	-	-	aromatic amino acid HAAP transporter
235	1.7	1636222	1635853	1636310	457	FTL_1705	-65	+	-	cell division protein
236	1.7	928835	928811	928877	66	FTL_0960	-63	-	-	soluble pyridine nucleotide transhydrogenase
237	1.69	1443110	1442658	1443197	539	FTL_1511	-41	-	-	glycerophosphoryl diester phosphodiesterase family protein
237	1.69	1443110	1442658	1443197	539	FTL_1512	343	+	-	hypothetical protein

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

239	1.68	1608756	1608628	1608929	301	FTL_1673	-84	+	-	major facilitator transporter
240	1.68	1200406	1200287	1200575	288	FTL_1257	-48	+	-	GTP cyclohydrolase I, pseudogene
240	1.68	1200406	1200287	1200575	288	FTL_1258	862	+	-	aldo/keto reductase
241	1.68	356185	355402	356930	1528	FTL_0388	862	+	-	cation transporter
242	1.67	1411685	1411552	1412481	929	FTL_1485	-86	+	-	hypothetical protein
243	1.67	1633624	1633329	1634167	838	FTL_1701	625	-	*glpX*	fructose 1,6-bisphosphatase II
243	1.67	1633624	1633329	1634167	838	FTL_1702	-33	-	-	hypothetical protein
244	1.67	1034224	1034124	1034392	268	FTL_1083	327	-	-	hypothetical protein
245	1.66	594638	594517	594728	211	FTL_0606	-51	+	-	dTDP-D-glucose 4,6-dehydratase
248	1.66	192469	192370	192637	267	FTL_0190	364	-	-	major facilitator transporter
248	1.66	192469	192370	192637	267	FTL_0191	-4	+	-	cytochrome O ubiquinol oxidase subunit II
248	1.66	192469	192370	192637	267	FTL_0192	937	+	-	cytochrome O ubiquinol oxidase subunit I
249	1.66	10057	10015	10101	86	FTL_0013	798	+	-	regulatory protein recX
250	1.66	1027926	1027809	1028116	307	FTL_1076	752	+	-	short chain dehydrogenase family protein
251	1.66	1510109	1509821	1510259	438	FTL_1582	-53	-	-	putrescine-binding periplasmic protein
251	1.66	1510109	1509821	1510259	438	FTL_1583	137	+	-	glutamate:gamma-aminobutyric acid antiporter family protein
252	1.66	668453	667585	669181	1596	FTL_0680	408	-	-	polyamine transporter, subunit H, ABC transporter, membrane protein
254	1.65	667435	667244	667568	324	FTL_0679	285	-	-	polyamine transporter, subunit I, ABC transporter, membrane protein
255	1.65	1472909	1472698	1473207	509	FTL_1542	-80	-	-	hypothetical protein
255	1.65	1472909	1472698	1473207	509	FTL_1543	173	+	-	formamidopyrimidine-DNA glycosylase

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

256	1.65	1440796	1440493	1440956	463	FTL_1508	879	-	-	major facilitator transporter, pseudogene
256	1.65	1440796	1440493	1440956	463	FTL_1509	118	-	-	D-alanyl-D-alanine carboxypeptidase/D-alanyl-D-alanine-endopeptidase
257	1.65	966581	966356	966930	574	FTL_1000	111	+	-	hypothetical protein
257	1.65	966581	966356	966930	574	FTL_1001	330	+	-	hypothetical protein
259	1.65	1199041	1198824	1199120	296	FTL_1252	917	-	-	ATP-dependent RNA helicase RhlE
259	1.65	1199041	1198824	1199120	296	FTL_1255	61	+	-	GTP cyclohydrolase I, pseudogene
260	1.64	706886	706826	707096	270	FTL_0715	620	-	*thyA*	thymidylate synthase
261	1.64	59061	58932	59209	277	FTL_0058	-35	-	-	aromatic amino acid HAAP transporter
261	1.64	59061	58932	59209	277	FTL_0059	135	+	*gidB*	16S rRNA methyltransferase GidB
261	1.64	59061	58932	59209	277	FTL_0060	749	+	-	hypothetical protein
262	1.64	571838	571642	572581	939	FTL_0590	799	+	-	ATp-dependent helicase
264	1.64	1639610	1639379	1639875	496	FTL_1708	375	+	-	potassium uptake protein
266	1.63	1345925	1345754	1346140	386	FTL_1416	-82	-	-	capsule biosynthesis protein capB
267	1.62	882555	882458	882675	217	FTL_0906	-80	+	*engB*	ribosome biogenesis GTP-binding protein YsxC
267	1.62	882555	882458	882675	217	FTL_0907	656	+	-	ribosome biogenesis GTP-binding protein YsxC, pseudogene
268	1.62	1529283	1529037	1529558	521	FTL_1599	-87	-	-	hypothetical protein
270	1.62	1757484	1757407	1757682	275	FTL_1822	821	-	-	NADH dehydrogenase subunit I
271	1.61	1294834	1294708	1294945	237	FTL_1361	-38	+	-	cold shock protein
272	1.61	1603602	1603333	1603749	416	FTL_1670	-52	+	-	disulfide bond formation protein
272	1.61	1603602	1603333	1603749	416	FTL_1671	452	+	-	RND efflux transporter
273	1.61	1130566	1130434	1131085	651	FTL_1177	409	-	*trmE*	tRNA modification GTPase

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

										TrmE
273	1.61	1130566	1130434	1131085	651	FTL_1178	158	-	-	hypothetical protein
273	1.61	1130566	1130434	1131085	651	FTL_1179	-39	+	-	sigma-54 modulation protein
273	1.61	1130566	1130434	1131085	651	FTL_1180	294	+	-	PEP-dependent sugar PTS system family protein
273	1.61	1130566	1130434	1131085	651	FTL_1181	742	+	-	SpoU rRNA methylase family protein
274	1.61	1621700	1621540	1621939	399	FTL_1688	269	-	-	transposase, pseudogene
276	1.6	957082	956975	957218	243	FTL_0986	-48	-	-	ribonucleotide-diphosphate reductase subunit alpha
277	1.6	351199	350990	351292	302	FTL_0378	709	-	-	hypothetical protein
277	1.6	351199	350990	351292	302	FTL_0379	178	-	-	methionine sulfoxide reductase B
277	1.6	351199	350990	351292	302	FTL_0380	-17	+	-	superoxide dismuate (Cu-Zn) precusor
277	1.6	351199	350990	351292	302	FTL_0381	544	+	-	hypothetical protein
278	1.6	1046702	1046201	1046990	789	FTL_1098	837	-	-	hypothetical protein, pseudogene
278	1.6	1046702	1046201	1046990	789	FTL_1099	281	-	-	hypothetical protein, pseudogene
278	1.6	1046702	1046201	1046990	789	FTL_1101	160	+	-	hypothetical protein
278	1.6	1046702	1046201	1046990	789	FTL_1102	522	+	-	hypothetical protein, pseudogene
278	1.6	1046702	1046201	1046990	789	FTL_1103	913	+	-	hypothetical protein, pseudogene
279	1.6	1451712	1451638	1451842	204	FTL_1521	61	-	-	chitinase family 18 protein
279	1.6	1451712	1451638	1451842	204	FTL_1522	473	+	-	2-amino-3-ketobutyrate coenzyme A ligase
280	1.59	311906	311564	312083	519	FTL_0325	245	-	-	OmpA family protein
280	1.59	311906	311564	312083	519	FTL_0326	-53	+	-	FKBP-type 16 kDa peptidyl-prolyl cis-transisomerase
280	1.59	311906	311564	312083	519	FTL_0327	392	+	*ispH*	4-hydroxy-3-methylbut-2-enyl diphosphate reductase
282	1.59	344335	344245	344399	154	FTL_0372	16	+	*gpsA*	NAD(P)H-dependent glycerol-

Table S3. Genes associated* with GreA (FTL_1474) ChIP-Seq peaks, continued

										3-phosphate dehydrogenase
283	1.59	455185	455043	455351	308	FTL_0477	-63	+	*gcvT*	glycine cleavage system aminomethyltransferase T
284	1.58	361058	360959	361258	299	FTL_0390	3	-	-	Type IV pili fiber building block protein
292	1.55	1600919	1600466	1601112	646	FTL_1667	431	-	*lpxK*	tetraacyldisaccharide 4'-kinase
293	1.55	864068	864005	864128	123	FTL_0884	841	-	-	hypothetical protein
295	1.55	1722053	1721993	1722126	133	FTL_1790	569	+	-	major facilitator superfamily transporter
297	1.53	671310	670845	671434	589	FTL_0682	981	-	*isftu1*	transposase
299	1.53	195053	195003	195119	116	FTL_0193	380	+	-	cytochrome O ubiquinol oxidase, subunit III
299	1.53	195053	195003	195119	116	FTL_0194	984	+	-	cytochrome O ubiquinol oxidase subunit IV
300	1.53	774896	774639	774941	302	FTL_0789	52	+	-	aspartate aminotransferase

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S4. Genes associated* with ParB (FTL_0428) ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	14.03	1885549	1876197	1895850	19653	FTL_1957	492	+	-	heat shock protein
4	2.29	1874909	1874356	1875318	962	FTL_1945	273	-	-	hypothetical protein
6	1.96	396361	396036	396566	530	FTL_0429	193	+	-	glutamine amidotransferase
6	1.96	396361	396036	396566	530	FTL_0430	903	+	-	hypothetical protein
7	1.95	1875933	1875366	1876132	766	FTL_1948	992	+	-	major facilitator transporter
8	1.85	1870172	1869873	1870922	1049	FTL_1940	877	-	-	trp repressor binding protein
8	1.85	1870172	1869873	1870922	1049	FTL_1942	336	+	-	transporter
8	1.85	1870172	1869873	1870922	1049	FTL_1943	908	+	-	primosomal protein N'
9	1.61	645155	645010	645298	288	FTL_0662	172	-	-	prophage repressor protein
9	1.61	645155	645010	645298	288	FTL_0663	-3	+	-	hypothetical protein

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S5. Genes associated* with LexA (FTL_0662) ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	14.59	645148	644822	646559	1737	FTL_0662	165	-	-	prophage repressor protein
1	14.59	645148	644822	646559	1737	FTL_0663	4	+	-	hypothetical protein
3	1.64	1738516	1738330	1738656	326	FTL_1804	998	-	-	ribosomal large subunit pseudouridine synthase C
3	1.64	1738516	1738330	1738656	326	FTL_1805	-62	-	-	ATPase
6	1.52	520033	519941	520304	363	FTL_0536	150	+	-	outer membrane protein OmpH
6	1.52	520033	519941	520304	363	FTL_0537	672	+	*lpxD*	UDP-3-O-[3-hydroxymyristoyl] glucosamine N-acyltransferase

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S6. Genes associated* with HipB (FTL_1126) ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	445.46	1071593	1070842	1072155	1313	FTL_1125	347	-	-	transcriptional regulator
1	445.46	1071593	1070842	1072155	1313	FTL_1126	117	-	-	transcriptional regulator
2	4.23	1894105	1893908	1895738	1830	FTL_1966	588	-	-	anthranilate synthase component I
2	4.23	1894105	1893908	1895738	1830	FTL_1967	292	-	-	trp operon repressor
2	4.23	1894105	1893908	1895738	1830	FTL_R0051	-40	-	*tRNA-Asp1*	Asp tRNA
3	3.55	1347764	1347206	1348114	908	FTL_1417	416	-	-	major facilitator transporter
3	3.55	1347764	1347206	1348114	908	FTL_R0035	166	-	*tRNA-Leu4*	Leu tRNA
3	3.55	1347764	1347206	1348114	908	FTL_R0036	84	-	*tRNA-His1*	His tRNA
3	3.55	1347764	1347206	1348114	908	FTL_R0037	-20	-	*tRNA-Arg2*	Arg tRNA
4	3.49	643973	643745	644356	611	FTL_R0025	-12	+	*tRNA-Cys1*	Cys tRNA
4	3.49	643973	643745	644356	611	FTL_R0026	67	+	*tRNA-Leu2*	Leu tRNA
5	3.36	1686559	1684198	1686925	2727	FTL_1751	274	-	-	elongation factor Tu
5	3.36	1686559	1684198	1686925	2727	FTL_R0046	154	-	*tRNA-Thr2*	Thr tRNA
5	3.36	1686559	1684198	1686925	2727	FTL_R0047	52	-	*tRNA-Gly2*	Gly tRNA
5	3.36	1686559	1684198	1686925	2727	FTL_R0048	-34	-	*tRNA-Tyr1*	Tyr tRNA
6	3.25	1293159	1292940	1293328	388	FTL_1358	864	-	-	cation-efflux family protein
6	3.25	1293159	1292940	1293328	388	FTL_1359	204	-	-	hypothetical protein
6	3.25	1293159	1292940	1293328	388	FTL_R0034	-60	+	*tRNA-Ser3*	Ser tRNA
8	2.99	125967	124375	129688	5313	FTL_R0004	327	+	*tRNA-Ile1*	Ile tRNA
8	2.99	125967	124375	129688	5313	FTL_R0005	418	+	*tRNA-Ala1*	Ala tRNA
8	2.99	125967	124375	129688	5313	FTL_R0006	548	+	-	23S ribosomal RNA
9	2.91	293270	292962	293660	698	FTL_0306	172	-	-	tryptophanyl-tRNA synthetase
9	2.91	293270	292962	293660	698	FTL_0308	937	+	-	hypothetical protein

Table S6. Genes associated* with HipB (FTL_1126) ChIP-Seq peaks, continued

9	2.91	293270	292962	293660	698	FTL_R0011	-2	-	*tRNA-Gly3*	Gly tRNA
9	2.91	293270	292962	293660	698	FTL_R0012	-93	-	*tRNA-Arg3*	Arg tRNA
10	2.85	223325	222971	223633	662	FTL_R0009	-3	+	*tRNA-Glu1*	Glu tRNA
11	2.64	1746270	1745972	1746435	463	FTL_1810	629	-	*nusA*	transcription elongation factor NusA
11	2.64	1746270	1745972	1746435	463	FTL_1811	159	-	-	hypothetical protein
11	2.64	1746270	1745972	1746435	463	FTL_R0050	7	-	*tRNA-Met2*	Met tRNA
12	2.63	636563	636286	636775	489	FTL_0652	424	-	-	hypothetical protein
12	2.63	636563	636286	636775	489	FTL_0653	283	+	-	hypothetical protein
12	2.63	636563	636286	636775	489	FTL_0654	700	+	-	uroporphyrinogen III synthase
12	2.63	636563	636286	636775	489	FTL_R0023	-56	-	*tRNA-Asn1*	Asn tRNA
13	2.52	414337	414083	414638	555	FTL_0444	236	-	*metG*	methionyl-tRNA synthetase
13	2.52	414337	414083	414638	555	FTL_R0014	-57	+	*tRNA-Ser2*	Ser tRNA
13	2.52	414337	414083	414638	555	FTL_R0015	62	+	*tRNA-Val1*	Val tRNA
14	2.48	1120683	1118962	1124304	5342	FTL_R0029	250	+	*tRNA-Ile3*	Ile tRNA
14	2.48	1120683	1118962	1124304	5342	FTL_R0030	341	+	*tRNA-Ala3*	Ala tRNA
14	2.48	1120683	1118962	1124304	5342	FTL_R0031	471	+	-	23S ribosomal RNA
17	2.11	1414393	1414110	1414569	459	FTL_R0040	-37	-	*tRNA-Lys1*	Lys tRNA
18	2.09	526959	526656	527119	463	FTL_R0021	-57	-	*tRNA-Met3*	Met tRNA
19	2.07	193164	192943	193366	423	FTL_0192	242	+	-	cytochrome O ubiquinol oxidase subunit I
20	2.03	1892600	1890379	1893712	3333	FTL_1965	631	-	-	hypothetical protein, pseudogene
21	1.99	158549	158282	158873	591	FTL_0152	143	+	-	hypothetical protein
21	1.99	158549	158282	158873	591	FTL_0153	487	+	*isftu1*	transposase
21	1.99	158549	158282	158873	591	FTL_R0008	-27	+	*tRNA-Gln1*	Gln tRNA
22	1.94	75742	75418	75839	421	FTL_0078	138	-	-	riboflavin biosynthesis protein ribD
23	1.92	1463193	1462996	1463515	519	FTL_1531	797	-	*rumA*	23S rRNA 5-methyluridine methyltransferase
23	1.92	1463193	1462996	1463515	519	FTL_1532	136	-	-	hypothetical protein

Table S6. Genes associated* with HipB (FTL_1126) ChIP-Seq peaks, continued

23	1.92	1463193	1462996	1463515	519	FTL_R0041	-64	-	*tRNA-Val3*	Val tRNA
24	1.92	1642909	1642499	1643283	784	FTL_1709	967	-	-	hypothetical protein
24	1.92	1642909	1642499	1643283	784	FTL_1710	186	-	-	ProP osmoprotectant transporter, fragment
24	1.92	1642909	1642499	1643283	784	FTL_R0042	-9	+	*tRNA-Met1*	Met tRNA
25	1.81	1002990	1002708	1003167	459	FTL_1046	288	-	-	D-alanyl-D-alanine carboxypeptidase
25	1.81	1002990	1002708	1003167	459	FTL_1047	-32	+	*rpsU*	30S ribosomal protein S21
25	1.81	1002990	1002708	1003167	459	FTL_1048	226	+	-	hypothetical protein
25	1.81	1002990	1002708	1003167	459	FTL_1049	693	+	-	DNA primase
26	1.55	1844325	1843833	1845158	1325	FTL_1910	305	-	-	D-alanyl-alanine synthetase A
26	1.55	1844325	1843833	1845158	1325	FTL_1912	308	+	*rpsA*	30S ribosomal protein S1

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S7. Genes associated* with ArsR (FTL_0370) ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	35.42	343312	342407	344713	2306	FTL_0368	923	-	-	pseudogene
1	35.42	343312	342407	344713	2306	FTL_0369	439	-	-	pseudogene
1	35.42	343312	342407	344713	2306	FTL_0370	143	-	-	arsenical restistance operon repressor
1	35.42	343312	342407	344713	2306	FTL_0371	66	+	-	hypothetical protein

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S8. Genes associated* with FTL_1293 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	1.79	1168046	1167730	1168925	1195	FTL_1217	821	-	-	hypothetical protein
2	1.7	764815	764315	765229	914	FTL_0777	259	+	-	hypothetical protein
2	1.7	764815	764315	765229	914	FTL_0778	706	+	-	hypothetical protein, pseudogene
3	1.69	947915	947676	947991	315	FTL_0977	607	+	-	hypothetical protein
5	1.65	1167137	1166585	1167592	1007	FTL_1216	917	-	-	hypothetical protein
6	1.65	943737	943517	944143	626	FTL_0974	168	+	-	50S ribosomal protein L11 methyltransferase
8	1.64	1236095	1235956	1236402	446	FTL_1295	842	-	-	amino acid transporter
8	1.64	1236095	1235956	1236402	446	FTL_1296	294	-	-	amino acid antiporter
9	1.64	1270515	1270163	1270955	792	FTL_1334	484	-	-	L-serine dehydratase 1
10	1.62	212761	211816	213380	1564	FTL_0212	-35	+	-	hypothetical protein
12	1.62	669290	669122	669349	227	FTL_0681	124	-	-	polyamine transporter, ABC transporter,ATP-binding protein
13	1.62	1067866	1067677	1068044	367	FTL_1123	600	+	-	hypothetical protein
14	1.62	1258912	1258690	1259676	986	FTL_1322	815	-	-	hypothetical protein, pseudogene
14	1.62	1258912	1258690	1259676	986	FTL_1323	132	-	-	hypothetical protein, pseudogene
14	1.62	1258912	1258690	1259676	986	FTL_1324	-10	-	-	hypothetical protein, pseudogene
15	1.61	52909	52648	53142	494	FTL_0050	740	-	-	hypothetical protein
15	1.61	52909	52648	53142	494	FTL_0051	194	-	-	hypothetical protein
17	1.6	1373635	1373448	1373796	348	FTL_1446	318	-	*isftu1*	transposase
18	1.6	1296150	1295983	1296414	431	FTL_1362	94	-	-	hypothetical protein
18	1.6	1296150	1295983	1296414	431	FTL_1363	112	+	-	hypothetical protein
19	1.6	822472	822315	822615	300	FTL_0838	900	-	-	D-methionine transport protein, ABC transporter,ATP-

Table S8. Genes associated* with FTL_1293 peaks, continued

										binding subunit
19	1.6	822472	822315	822615	300	FTL_0839	299	-	-	hypothetical protein
19	1.6	822472	822315	822615	300	FTL_0840	337	+	-	hypothetical protein
19	1.6	822472	822315	822615	300	FTL_0841	669	+	-	lipoprotein
20	1.6	1353071	1352681	1353562	881	FTL_1421	901	-	-	haloacid dehalogenase-like hydrolase family protein
21	1.59	445729	443800	445946	2146	FTL_0472	199	+	-	DNA polymerase III subunit alpha
22	1.59	1365867	1365531	1366216	685	FTL_1434	314	-	-	DoxD-like family protein
23	1.59	343331	343059	343647	588	FTL_0368	942	-	-	hypothetical protein, pseudogene
23	1.59	343331	343059	343647	588	FTL_0369	458	-	-	hypothetical protein, pseudogene
23	1.59	343331	343059	343647	588	FTL_0370	162	-	-	arsenical restistance operon repressor
23	1.59	343331	343059	343647	588	FTL_0371	47	+	-	hypothetical protein
24	1.58	1314855	1314753	1315390	637	FTL_1382	852	-	-	hypothetical protein, pseudogene
24	1.58	1314855	1314753	1315390	637	FTL_1383	340	-	-	glutathione peroxidase
24	1.58	1314855	1314753	1315390	637	FTL_1384	-36	-	-	hypothetical protein
24	1.58	1314855	1314753	1315390	637	FTL_1385	133	+	-	hypothetical protein
25	1.58	859149	858549	859258	709	FTL_0879	961	-	-	beta-lactamase
26	1.57	114672	114018	115397	1379	FTL_0119	708	-	-	hypothetical protein
27	1.57	1879012	1877525	1879342	1817	FTL_1949	290	-	-	hypothetical protein
28	1.57	949427	948992	949964	972	FTL_0980	190	+	-	hypothetical protein
29	1.57	32101	31918	32447	529	FTL_0030	212	-	-	carbamoyl phosphate synthase small subunit
29	1.57	32101	31918	32447	529	FTL_0031	205	+	-	acid phosphatase
30	1.56	1031196	1030992	1031421	429	FTL_1078	54	-	-	hypothetical protein, pseudogene
32	1.56	1102939	1102149	1103540	1391	FTL_1161	915	-	-	hypothetical protein
33	1.56	143218	142565	143390	825	FTL_0136	775	-	-	hypothetical protein
34	1.56	36624	35513	37342	1829	FTL_0034	-87	-	-	hypothetical protein

Table S8. Genes associated* with FTL_1293 peaks, continued

34	1.56	36624	35513	37342	1829	FTL_0035	624	+	-	hypothetical protein
35	1.56	477509	477279	477790	511	FTL_0492	201	+	-	UDP-N--acetylmuramoylalanyl-D-glutamyl-2,6- diaminopimelate-D-alanyl-D-alanyl ligase
36	1.56	1556331	1555716	1556536	820	FTL_1627	322	+	*isftu1*	transposase
37	1.55	1489960	1489694	1490371	677	FTL_1557	465	-	-	hypothetical protein
37	1.55	1489960	1489694	1490371	677	FTL_1558	-68	-	-	hypothetical protein
37	1.55	1489960	1489694	1490371	677	FTL_1559	292	+	-	hypothetical protein, pseudogene
39	1.54	443638	443558	443713	155	FTL_0467	800	-	-	hypothetical protein
39	1.54	443638	443558	443713	155	FTL_0468	196	-	*isftu2*	transposase
39	1.54	443638	443558	443713	155	FTL_0470	780	+	-	hypothetical protein
40	1.54	465778	465627	465950	323	FTL_0483	98	-	-	glycogen branching protein
40	1.54	465778	465627	465950	323	FTL_0484	203	+	-	phosphoglucomutase
41	1.54	645253	644956	645577	621	FTL_0662	270	-	-	prophage repressor protein
42	1.54	1262836	1262755	1263336	581	FTL_1326	433	-	-	hypothetical protein
43	1.54	1573059	1572701	1573237	536	FTL_1643	298	+	*isftu1*	transposase
44	1.53	1281321	1280868	1282403	1535	FTL_1347	-87	-	-	hypothetical protein
44	1.53	1281321	1280868	1282403	1535	FTL_1348	380	+	-	transposase
48	1.51	1066776	1066731	1066854	123	FTL_1121	-4	+	-	hypothetical protein
49	1.51	1376788	1376239	1376926	687	FTL_1450	118	+	-	major facilitator transporter

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S9. Genes associated* with BirA (FTL_1276) ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	1.83	692270	692219	692337	118	FTL_0702	253	-	-	hypothetical protein
2	1.83	669288	669096	669354	258	FTL_0681	122	-	-	polyamine transporter, ABC transporter,ATP-binding protein
3	1.81	1059218	1058948	1059480	532	FTL_1113	310	-	-	hypothetical protein
3	1.81	1059218	1058948	1059480	532	FTL_1115	816	+	-	hypothetical protein
4	1.81	1881691	1881209	1881953	744	FTL_1954	450	+	-	hypothetical protein
5	1.8	162424	161896	162526	630	FTL_0156	671	-	-	phosphate transport protein
5	1.8	162424	161896	162526	630	FTL_0157	-3	-	-	hypothetical protein
5	1.8	162424	161896	162526	630	FTL_0158	310	+	-	acid phosphatase
6	1.79	1305897	1305739	1306007	268	FTL_1372	214	-	-	lipoprotein
6	1.79	1305897	1305739	1306007	268	FTL_1373	259	+	-	hypothetical protein
6	1.79	1305897	1305739	1306007	268	FTL_1374	964	+	-	organic solvent tolerance protein
7	1.79	343379	343109	343635	526	FTL_0368	990	-	-	Ser tRNA, pseudogene
7	1.79	343379	343109	343635	526	FTL_0369	506	-	-	Ser tRNA, pseudogene
7	1.79	343379	343109	343635	526	FTL_0370	210	-	-	arsenical restistance operon repressor
7	1.79	343379	343109	343635	526	FTL_0371	-1	+	-	hypothetical protein
7	1.79	343379	343109	343635	526	FTL_0372	972	+	*gpsA*	NAD(P)H-dependent glycerol-3-phosphate dehydrogenase
8	1.78	1003339	1003171	1003399	228	FTL_1046	637	-	-	D-alanyl-D-alanine carboxypeptidase
8	1.78	1003339	1003171	1003399	228	FTL_1049	344	+	-	DNA primase
9	1.76	879997	879911	880122	211	FTL_0901	349	-	-	monooxygenase family protein
9	1.76	879997	879911	880122	211	FTL_0902	185	-	-	oxidoreductase

Table S9. Genes associated* with BirA (FTL_1276) ChIP-Seq peaks, continued

9	1.76	879997	879911	880122	211	FTL_0903	0	+	-	hypothetical protein
10	1.75	470533	470268	470644	376	FTL_0487	-29	+	-	maltodextrin phosphorylase
11	1.73	1895011	1894229	1895773	1544	FTL_R0051	866	-	*tRNA-Asp1*	Asp tRNA
11	1.73	1895011	1894229	1895773	1544	FTL_R0052	786	-	*tRNA-Thr1*	Thr tRNA
13	1.73	995847	995584	996408	824	FTL_1039	566	+	-	hypothetical protein
13	1.73	995847	995584	996408	824	FTL_R0027	407	+	-	-
14	1.72	719719	719625	719985	360	FTL_0730	123	+	-	haloacid dehalogenase-like hydrolase family protein
15	1.71	822508	822352	822622	270	FTL_0838	936	-	-	D-methionine transport protein, ABC transporter,ATP-binding subunit
15	1.71	822508	822352	822622	270	FTL_0839	335	-	-	hypothetical protein
15	1.71	822508	822352	822622	270	FTL_0840	301	+	-	hypothetical protein
15	1.71	822508	822352	822622	270	FTL_0841	633	+	-	lipoprotein
16	1.7	143177	142572	143338	766	FTL_0136	734	-	-	hypothetical protein
18	1.68	1166966	1166271	1167631	1360	FTL_1216	746	-	-	hypothetical protein
20	1.67	1546434	1546139	1546520	381	FTL_1616	192	-	-	phosphoenolpyruvate carboxykinase
20	1.67	1546434	1546139	1546520	381	FTL_1617	-41	+	-	glutaminyl-tRNA synthetase
21	1.66	1888844	1885019	1890325	5306	FTL_1959	746	-	-	hypothetical protein
22	1.66	859154	858599	859275	676	FTL_0879	966	-	-	beta-lactamase
23	1.65	1738522	1738277	1738699	422	FTL_1805	-56	-	-	ATPase
24	1.65	949230	949028	949928	900	FTL_0979	67	+	-	hypothetical protein
24	1.65	949230	949028	949928	900	FTL_0980	387	+	-	hypothetical protein
25	1.64	1571054	1570273	1571346	1073	FTL_1640	263	+	-	amino acid transporter protein, fragment
26	1.64	1308739	1308655	1308964	309	FTL_1375	166	+	-	heat shock protein 15 (HSP15)
27	1.64	693735	693503	694671	1168	FTL_0703	187	-	*glyA*	serine hydroxymethyltransferase
27	1.64	693735	693503	694671	1168	FTL_0704	-83	+	-	hypothetical protein

Table S9. Genes associated* with BirA (FTL_1276) ChIP-Seq peaks, continued

28	1.64	783867	783721	784076	355	FTL_0799	-1	+	-	Type IV pili lipoprotein
28	1.64	783867	783721	784076	355	FTL_0800	602	+	-	Type IV pilin multimeric outer membrane protein
29	1.63	32097	31797	32189	392	FTL_0030	208	-	-	carbamoyl phosphate synthase small subunit
29	1.63	32097	31797	32189	392	FTL_0031	209	+	-	acid phosphatase
30	1.63	1321634	1321511	1321753	242	FTL_1390	723	-	-	quinolinate synthetase
30	1.63	1321634	1321511	1321753	242	FTL_1391	130	-	*gmk*	guanylate kinase
31	1.63	440603	440537	440853	316	FTL_0465	223	-	-	BNR/Asp-box repeat-containing protein
31	1.63	440603	440537	440853	316	FTL_0466	-24	+	-	soluble lytic murein transglycosylase
32	1.63	328237	327949	328424	475	FTL_0348	25	+	-	Ser tRNA, pseudogene
32	1.63	328237	327949	328424	475	FTL_0349	661	+	-	Ser tRNA, pseudogene
34	1.62	844096	843781	844284	503	FTL_0863	89	-	-	Sodium-dicarboxylate symporter family protein
34	1.62	844096	843781	844284	503	FTL_0864	113	+	-	SIS domain-containing protein
35	1.62	764869	764437	765267	830	FTL_0777	205	+	-	hypothetical protein
35	1.62	764869	764437	765267	830	FTL_0778	652	+	-	hypothetical protein, pseudogene
36	1.62	724101	723759	724367	608	FTL_0735	975	+	-	haloacid dehalogenase-like hydrolase family protein, pseudogene
37	1.62	913972	913704	914070	366	FTL_0946	914	+	-	major facilitator transporter
38	1.62	1356306	1355412	1356633	1221	FTL_1425	348	-	-	hypothetical protein
39	1.61	957290	957115	957350	235	FTL_0986	160	-	-	ribonucleotide-diphosphate reductase subunit alpha
40	1.61	445640	445064	445953	889	FTL_0472	288	+	-	DNA polymerase III subunit alpha
41	1.61	1296177	1295989	1296398	409	FTL_1362	121	-	-	hypothetical protein
41	1.61	1296177	1295989	1296398	409	FTL_1363	85	+	-	hypothetical protein
42	1.6	1316464	1316381	1316848	467	FTL_1386	199	-	*isftu2*	transposase

Table S9. Genes associated* with BirA (FTL_1276) ChIP-Seq peaks, continued

43	1.6	1365994	1365543	1366194	651	FTL_1434	441	-	-	DoxD-like family protein
43	1.6	1365994	1365543	1366194	651	FTL_1435	8	-	-	hypothetical protein
44	1.6	897643	897432	897763	331	FTL_0922	796	-	-	Iron-sulfur cluster-binding protein
44	1.6	897643	897432	897763	331	FTL_0923	150	-	-	glutaredoxin
44	1.6	897643	897432	897763	331	FTL_0924	-67	+	-	proton-dependent oligopeptide transporter
44	1.6	897643	897432	897763	331	FTL_0925	259	+	-	proton-dependent oligopeptide transport (POT) family protein
45	1.6	326384	326121	326904	783	FTL_0346	-42	+	-	hypothetical protein
45	1.6	326384	326121	326904	783	FTL_0347	585	+	-	hypothetical protein
46	1.59	1281053	1280845	1281224	379	FTL_1348	648	+	-	transposase
47	1.59	581575	581123	582165	1042	FTL_0596	904	+	-	UDP-glucose/GDP-mannose dehydrogenase
48	1.59	426299	425844	426437	593	FTL_0450	115	-	*psd*	phosphatidylserine decarboxylase
48	1.59	426299	425844	426437	593	FTL_0451	-16	+	-	hypothetical protein
48	1.59	426299	425844	426437	593	FTL_0452	973	+	-	bifunctional nicotinamide mononucleotide adenylyltransferase/ADP-ribose pyrophosphatase
49	1.58	1336052	1335972	1336175	203	FTL_1407	298	-	*thrS*	threonyl-tRNA synthetase
49	1.58	1336052	1335972	1336175	203	FTL_1408	208	+	-	chitin binding protein
50	1.58	681767	681512	682023	511	FTL_0691	949	-	-	proton-dependent oligopeptide transport (POT) family protein
50	1.58	681767	681512	682023	511	FTL_0692	-15	-	-	16S ribosomal RNA methyltransferase RsmE
50	1.58	681767	681512	682023	511	FTL_0693	86	+	-	hypothetical protein
51	1.58	1259250	1259110	1259540	430	FTL_1323	470	-	-	aldo/keto reductase, pseudogene
51	1.58	1259250	1259110	1259540	430	FTL_1324	328	-	-	aldo/keto reductase, pseudogene
52	1.57	1201092	1201004	1201193	189	FTL_1258	176	+	-	aldo/keto reductase

Table S9. Genes associated* with BirA (FTL_1276) ChIP-Seq peaks, continued

53	1.57	1270587	1270202	1270972	770	FTL_1334	556	-	-	L-serine dehydratase 1
54	1.57	52958	52720	53120	400	FTL_0050	789	-	-	hypothetical protein
54	1.57	52958	52720	53120	400	FTL_0051	243	-	-	hypothetical protein
55	1.56	1056925	1056820	1057012	192	FTL_1112	485	+	*isftu2*	transposase
56	1.56	1010799	1010534	1010899	365	FTL_1054	959	-	-	putative alpha-xylosidase
56	1.56	1010799	1010534	1010899	365	FTL_1055	65	-	-	putative alpha-xylosidase, pseudogene
57	1.55	1292964	1292897	1293433	536	FTL_1358	669	-	-	cation-efflux family protein
57	1.55	1292964	1292897	1293433	536	FTL_1359	9	-	-	hypothetical protein
57	1.55	1292964	1292897	1293433	536	FTL_R0034	135	+	*tRNA-Ser3*	Ser tRNA
58	1.55	371458	371194	371884	690	FTL_0400	388	-	*isftu2*	transposase
58	1.55	371458	371194	371884	690	FTL_0402	246	+	-	intracellular septation protein A family protein
58	1.55	371458	371194	371884	690	FTL_0403	850	+	-	hypothetical protein
59	1.55	672515	672321	672621	300	FTL_0684	994	-	-	hypothetical protein
59	1.55	672515	672321	672621	300	FTL_0685	141	-	-	NH(3)-dependent NAD(+) synthetase
60	1.54	935681	935488	935758	270	FTL_0967	125	-	-	lipoate-protein ligase A
60	1.54	935681	935488	935758	270	FTL_0968	-11	+	-	tyrosyl-tRNA synthetase
61	1.54	985676	985527	985742	215	FTL_1028	-59	+	-	radical SAM superfamily protein
63	1.54	958421	958147	958633	486	FTL_0987	128	-	-	malate dehydrogenase
63	1.54	958421	958147	958633	486	FTL_0988	35	+	-	hypothetical protein
63	1.54	958421	958147	958633	486	FTL_0989	563	+	-	ubiquinone biosynthesis protein
64	1.52	1384218	1384119	1384306	187	FTL_1458	-45	-	-	preprotein translocase subunit SecA
64	1.52	1384218	1384119	1384306	187	FTL_1459	177	+	-	hypothetical protein
65	1.52	738860	738674	739016	342	FTL_0748	75	+	-	major facilitator transporter
66	1.52	398762	398597	398916	319	FTL_0434	808	+	-	hypothetical protein
67	1.51	1727987	1727908	1728037	129	FTL_1793	-10	-	-	hypothetical protein

Table S9. Genes associated* with BirA (FTL_1276) ChIP-Seq peaks, continued

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S10. Genes associated* with FTL_1231 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	3.88	1120758	1118933	1124367	5434	FTL_R0029	175	+	*tRNA-Ile3*	Ile tRNA
1	3.88	1120758	1118933	1124367	5434	FTL_R0030	266	+	*tRNA-Ala3*	Ala tRNA
1	3.88	1120758	1118933	1124367	5434	FTL_R0031	396	+	-	23S ribosomal RNA
2	3.61	416963	415431	420810	5379	FTL_R0017	352	+	*tRNA-Ile2*	Ile tRNA
2	3.61	416963	415431	420810	5379	FTL_R0018	443	+	*tRNA-Ala2*	Ala tRNA
2	3.61	416963	415431	420810	5379	FTL_R0019	573	+	-	23S ribosomal RNA
3	3.54	126351	124298	129745	5447	FTL_R0004	-57	+	*tRNA-Ile1*	Ile tRNA
3	3.54	126351	124298	129745	5447	FTL_R0005	34	+	*tRNA-Ala1*	Ala tRNA
3	3.54	126351	124298	129745	5447	FTL_R0006	164	+	-	23S ribosomal RNA
4	3.29	1347815	1347309	1348165	856	FTL_1417	467	-	-	major facilitator transporter
4	3.29	1347815	1347309	1348165	856	FTL_R0035	217	-	*tRNA-Leu4*	Leu tRNA
4	3.29	1347815	1347309	1348165	856	FTL_R0036	135	-	*tRNA-His1*	His tRNA
4	3.29	1347815	1347309	1348165	856	FTL_R0037	31	-	*tRNA-Arg2*	Arg tRNA
4	3.29	1347815	1347309	1348165	856	FTL_R0038	-54	-	*tRNA-Pro1*	Pro tRNA
5	3.28	644041	643702	644273	571	FTL_R0025	-80	+	*tRNA-Cys1*	Cys tRNA
5	3.28	644041	643702	644273	571	FTL_R0026	-1	+	*tRNA-Leu2*	Leu tRNA
6	2.46	1894130	1893876	1894467	591	FTL_1966	613	-	-	anthranilate synthase component I
6	2.46	1894130	1893876	1894467	591	FTL_1967	317	-	-	trp operon repressor
6	2.46	1894130	1893876	1894467	591	FTL_R0051	-15	-	*tRNA-Asp1*	Asp tRNA
6	2.46	1894130	1893876	1894467	591	FTL_R0052	-95	-	*tRNA-Thr1*	Thr tRNA
7	2.31	1686473	1685489	1686831	1342	FTL_1751	188	-	-	elongation factor Tu
7	2.31	1686473	1685489	1686831	1342	FTL_R0046	68	-	*tRNA-Thr2*	Thr tRNA
7	2.31	1686473	1685489	1686831	1342	FTL_R0047	-34	-	*tRNA-Gly2*	Gly tRNA
8	2.13	1796033	1795992	1796070	78	FTL_1864	407	-	-	hypothetical protein
9	1.89	1441551	1441511	1441625	114	FTL_1509	873	-	-	D-alanyl-D-alanine

Table S10. Genes associated* with FTL_1231 ChIP-Seq peaks, continued

										carboxypeptidase/D-alanyl-D-alanine-endopeptidase
10	1.88	293308	292931	293999	1068	FTL_0306	210	-	-	tryptophanyl-tRNA synthetase
10	1.88	293308	292931	293999	1068	FTL_0308	899	+	-	hypothetical protein
10	1.88	293308	292931	293999	1068	FTL_R0011	36	-	*tRNA-Gly3*	Gly tRNA
10	1.88	293308	292931	293999	1068	FTL_R0012	-55	-	*tRNA-Arg3*	Arg tRNA
11	1.83	414316	413925	414725	800	FTL_0444	215	-	*metG*	methionyl-tRNA synthetase
11	1.83	414316	413925	414725	800	FTL_R0014	-36	+	*tRNA-Ser2*	Ser tRNA
11	1.83	414316	413925	414725	800	FTL_R0015	83	+	*tRNA-Val1*	Val tRNA
12	1.83	223363	223065	223676	611	FTL_R0009	-41	+	*tRNA-Glu1*	Glu tRNA
13	1.82	636559	636336	636857	521	FTL_0652	420	-	-	hypothetical protein
13	1.82	636559	636336	636857	521	FTL_0653	287	+	-	hypothetical protein
13	1.82	636559	636336	636857	521	FTL_0654	704	+	-	uroporphyrinogen III synthase
13	1.82	636559	636336	636857	521	FTL_R0023	-60	-	*tRNA-Asn1*	Asn tRNA
15	1.79	1293144	1292828	1293326	498	FTL_1358	849	-	-	cation-efflux family protein
15	1.79	1293144	1292828	1293326	498	FTL_1359	189	-	-	hypothetical protein
15	1.79	1293144	1292828	1293326	498	FTL_R0034	-45	+	*tRNA-Ser3*	Ser tRNA
16	1.77	10129	9880	10205	325	FTL_0013	726	+	-	regulatory protein recX
18	1.75	1440857	1440667	1441005	338	FTL_1508	940	-	-	Phe tRNA, pseudogene
20	1.73	315250	315188	315330	142	FTL_0330	591	-	-	hypothetical protein
20	1.73	315250	315188	315330	142	FTL_0332	302	+	-	TolR protein
20	1.73	315250	315188	315330	142	FTL_0333	729	+	-	hypothetical protein
21	1.72	1746152	1745907	1746463	556	FTL_1810	511	-	*nusA*	transcription elongation factor NusA
21	1.72	1746152	1745907	1746463	556	FTL_1811	41	-	-	hypothetical protein
22	1.69	1734553	1733899	1735015	1116	FTL_1800	673	-	-	F0F1 ATP synthase subunit C
22	1.69	1734553	1733899	1735015	1116	FTL_1803	730	+	-	major facilitator transporter
24	1.68	1449535	1449450	1449594	144	FTL_1520	370	-	*upp*	uracil phosphoribosyltransferase
25	1.67	266250	266168	266312	144	FTL_0280	374	-	-	amino acid permease

Table S10. Genes associated* with FTL_1231 ChIP-Seq peaks, continued

26	1.67	1667145	1666753	1667401	648	FTL_1734	897	-	-	integrase/recombinase
26	1.67	1667145	1666753	1667401	648	FTL_1735	384	-	*rplS*	50S ribosomal protein L19
27	1.67	52288	52226	52357	131	FTL_0050	119	-	-	hypothetical protein
29	1.64	233409	233357	233701	344	FTL_0235	582	+	*rpsJ*	30S ribosomal protein S10
31	1.64	230548	230522	230763	241	FTL_0232	407	+	*rpsL*	30S ribosomal protein S12
32	1.63	1869130	1869058	1869231	173	FTL_1939	442	-	-	outer membrane lipoprotein
32	1.63	1869130	1869058	1869231	173	FTL_1941	172	+	-	tRNA processing ribonuclease BN
35	1.62	151383	151323	151474	151	FTL_0146	265	+	-	ABC transporter ATP-binding protein
37	1.61	1710628	1710203	1710930	727	FTL_1778	613	-	-	Tyr tRNA, pseudogene
37	1.61	1710628	1710203	1710930	727	FTL_1779	-47	-	*secG*	preprotein translocase subunit SecG
37	1.61	1710628	1710203	1710930	727	FTL_R0049	315	-	*tRNA-Leu3*	Leu tRNA
39	1.6	30866	30808	30911	103	FTL_0029	148	-	*carB*	carbamoyl phosphate synthase large subunit
42	1.56	1602105	1602022	1602306	284	FTL_1669	323	+	-	tRNA CCA-pyrophosphorylase
43	1.55	1333901	1333336	1334190	854	FTL_1404	870	-	*rplT*	50S ribosomal protein L20
43	1.55	1333901	1333336	1334190	854	FTL_1405	637	-	*rpmI*	50S ribosomal protein L35
43	1.55	1333901	1333336	1334190	854	FTL_1406	127	-	-	translation initiation factor IF-3
44	1.55	1892169	1890424	1893066	2642	FTL_1964	775	-	-	tRNA processing ribonuclease BN, pseudogene
44	1.55	1892169	1890424	1893066	2642	FTL_1965	200	-	-	tRNA processing ribonuclease BN, pseudogene
46	1.52	405951	405841	406034	193	FTL_0439	501	+	-	hypothetical protein

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

Table S10. Genes associated* with FTL_1231 ChIP-Seq peaks, continued

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S11. Genes associated* with FTL_1079 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	1.75	1032664	1032329	1033401	1072	FTL_1079	665	-	-	helix-turn-helix family protein
1	1.75	1032664	1032329	1033401	1072	FTL_1080	115	-	-	hypothetical protein
1	1.75	1032664	1032329	1033401	1072	FTL_1081	238	+	-	hypothetical protein
1	1.75	1032664	1032329	1033401	1072	FTL_1082	626	+	-	hypothetical protein
2	1.7	1280960	1280834	1282397	1563	FTL_1348	741	+	-	transposase
3	1.67	822474	822367	822618	251	FTL_0838	902	-	-	D-methionine transport protein, ABC transporter,ATP-binding subunit
3	1.67	822474	822367	822618	251	FTL_0839	301	-	-	hypothetical protein
3	1.67	822474	822367	822618	251	FTL_0840	335	+	-	hypothetical protein
3	1.67	822474	822367	822618	251	FTL_0841	667	+	-	lipoprotein
4	1.65	695270	694902	695496	594	FTL_0706	424	+	-	hypothetical protein
5	1.64	1262875	1262775	1264484	1709	FTL_1326	472	-	-	hypothetical protein
7	1.62	764900	764309	765288	979	FTL_0777	174	+	-	hypothetical protein
7	1.62	764900	764309	765288	979	FTL_0778	621	+	-	hypothetical protein, pseudogene
8	1.61	717981	717743	718288	545	FTL_0729	210	+	*queA*	S-adenosylmethionine--tRNA ribosyltransferase-isomerase
9	1.61	1365995	1365543	1366226	683	FTL_1434	442	-	-	DoxD-like family protein
9	1.61	1365995	1365543	1366226	683	FTL_1435	9	-	-	hypothetical protein
10	1.6	949575	948960	949934	974	FTL_0980	42	+	-	hypothetical protein
11	1.59	1236152	1235950	1236448	498	FTL_1295	899	-	-	amino acid transporter
11	1.59	1236152	1235950	1236448	498	FTL_1296	351	-	-	amino acid antiporter
11	1.59	1236152	1235950	1236448	498	FTL_1298	947	+	*isftu1*	transposase
12	1.59	1166988	1166229	1169462	3233	FTL_1216	768	-	-	hypothetical protein
13	1.59	645302	644899	646516	1617	FTL_0662	319	-	-	prophage repressor protein
15	1.55	1270395	1270201	1271024	823	FTL_1334	364	-	-	L-serine dehydratase 1

Table S11. Genes associated* with FTL_1079 ChIP-Seq peaks, continued

16	1.55	326255	326074	326885	811	FTL_0346	87	+	-	hypothetical protein
16	1.55	326255	326074	326885	811	FTL_0347	714	+	-	hypothetical protein
17	1.54	615434	615305	615642	337	FTL_0625	25	-	-	glycosyl transferase family protein
17	1.54	615434	615305	615642	337	FTL_0626	166	+	-	inorganic phosphate/ATP-NAD kinase

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S12. Genes associated* with FTL_1176 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	1.87	880029	879937	880098	161	FTL_0901	381	-	-	monooxygenase family protein
1	1.87	880029	879937	880098	161	FTL_0902	217	-	-	oxidoreductase
1	1.87	880029	879937	880098	161	FTL_0903	-32	+	-	hypothetical protein
2	1.77	406406	406185	406485	300	FTL_0439	46	+	-	hypothetical protein
3	1.68	1881700	1880455	1882472	2017	FTL_1954	441	+	-	hypothetical protein
4	1.66	1003239	1003078	1003390	312	FTL_1046	537	-	-	D-alanyl-D-alanine carboxypeptidase
4	1.66	1003239	1003078	1003390	312	FTL_1048	-23	+	-	hypothetical protein
4	1.66	1003239	1003078	1003390	312	FTL_1049	444	+	-	DNA primase
6	1.57	470543	470474	470640	166	FTL_0487	-39	+	-	maltodextrin phosphorylase
7	1.57	1276004	1275946	1276075	129	FTL_1339	33	-	-	proton-dependent oligopeptide transport (POT) family protein

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S13. Genes associated* with FTL_1222 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	1125.56	1172749	1171610	1173560	1950	FTL_1221	-31	+	-	hypothetical protein
1	1125.56	1172749	1171610	1173560	1950	FTL_1222	578	+	-	hypothetical protein
2	608.32	1159188	1158233	1160409	2176	FTL_1209	344	-	-	hypothetical protein

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S14. Genes associated* with FTL_0742 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	1.66	223422	222983	223599	616	FTL_R0009	-100	+	*tRNA-Glu1*	Glu tRNA
2	1.58	1003166	1002846	1003388	542	FTL_1046	464	-	-	D-alanyl-D-alanine carboxypeptidase
2	1.58	1003166	1002846	1003388	542	FTL_1048	50	+	-	hypothetical protein
2	1.58	1003166	1002846	1003388	542	FTL_1049	517	+	-	DNA primase
3	1.54	919710	919660	919788	128	FTL_0950	-21	+	-	50S ribosomal protein L25
3	1.54	919710	919660	919788	128	FTL_0951	365	+	-	hypothetical protein

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S15. Genes associated* with FTL_1193 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
2	1.6	1003361	1003330	1003390	60	FTL_1046	659	-	-	D-alanyl-D-alanine carboxypeptidase
2	1.6	1003361	1003330	1003390	60	FTL_1049	322	+	-	DNA primase

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S16. Genes associated* with FTL_0062 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	1.53	1768759	1768425	1769064	639	FTL_1834	-54	+	-	diaminopimelate decarboxylase

*Gene are referred to as associated if the annotated translational start site is within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within 100 bp upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	535.8	692075	691443	692811	1368	FTL_0702	58	-	-	hypothetical protein
2	317.05	158767	158096	159073	977	FTL_0152	-75	+	-	hypothetical protein
2	317.05	158767	158096	159073	977	FTL_0153	269	+	*isftu1*	transposase
2	317.05	158767	158096	159073	977	FTL_R0008	-245	+	*tRNA-Gln1*	Gln tRNA
3	184.3	444269	443853	444859	1006	FTL_0468	827	-	*isftu2*	transposase
3	184.3	444269	443853	444859	1006	FTL_0469	204	-	-	hypothetical protein
3	184.3	444269	443853	444859	1006	FTL_0470	149	+	-	hypothetical protein
3	184.3	444269	443853	444859	1006	FTL_0471	889	+	-	hypothetical protein
4	173.1	822736	822353	823312	959	FTL_0839	563	-	-	hypothetical protein
4	173.1	822736	822353	823312	959	FTL_0840	73	+	-	hypothetical protein
4	173.1	822736	822353	823312	959	FTL_0841	405	+	-	lipoprotein
5	152.61	801439	801022	802056	1034	FTL_0814	934	-	-	hypothetical protein
5	152.61	801439	801022	802056	1034	FTL_0815	520	-	-	PRC-barrel protein
5	152.61	801439	801022	802056	1034	FTL_0816	268	-	-	hypothetical protein
6	149.97	485380	484988	485726	738	FTL_0499	-709	+	-	S-adenosylmethionine decarboxylase
6	149.97	485380	484988	485726	738	FTL_0500	-221	+	-	spermidine synthase
7	148.95	1336052	1335557	1336395	838	FTL_1407	298	-	*thrS*	threonyl-tRNA synthetase
7	148.95	1336052	1335557	1336395	838	FTL_1408	208	+	-	chitin binding protein
8	121.04	586103	585878	586570	692	FTL_0599	-100	+	-	glycosyl transferase family protein
8	121.04	586103	585878	586570	692	FTL_0600	1000	+	-	asparagine synthase
9	91.51	36157	35475	36509	1034	FTL_0034	-554	-	-	hypothetical protein
10	87.34	1451811	1451476	1452193	717	FTL_1521	160	-	-	chitinase family 18 protein
10	87.34	1451811	1451476	1452193	717	FTL_1522	374	+	-	2-amino-3-ketobutyrate coenzyme A ligase

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

11	60.25	1331844	1331614	1332238	624	FTL_1402	207	-	*isftu1*	transposase
11	60.25	1331844	1331614	1332238	624	FTL_1403	-720	-	-	hypothetical protein
12	48.88	50675	50327	51022	695	FTL_0047	806	-	-	hypothetical protein
12	48.88	50675	50327	51022	695	FTL_0048	140	-	-	prephenate dehydrogenase.
12	48.88	50675	50327	51022	695	FTL_0049	-565	-	-	D-tyrosyl-tRNA(Tyr) deacylase
13	47.46	450658	450408	450943	535	FTL_0474	123	+	-	lipoprotein releasing system, subunit C,putative membrane protein
13	47.46	450658	450408	450943	535	FTL_R0053	-479	+	-	5S ribosomal RNA
14	43.81	216552	216305	216806	501	FTL_0215	517	-	-	dihydropteridine reductase
14	43.81	216552	216305	216806	501	FTL_0216	-377	+	-	MutT/nudix family protein
14	43.81	216552	216305	216806	501	FTL_0217	173	+	*fumC*	fumarate hydratase
15	40.15	1564847	1564440	1565216	776	FTL_1634	507	-	-	LysR family transcriptional regulator
15	40.15	1564847	1564440	1565216	776	FTL_1635	31	+	-	chitinase, fragment
16	38.05	525455	525168	525834	666	FTL_0542	-570	+	-	hypothetical protein
16	38.05	525455	525168	525834	666	FTL_0543	211	+	-	hypothetical protein, pseudogene
17	31.41	1548559	1548323	1548848	525	FTL_1618	-499	+	-	hypothetical protein
17	31.41	1548559	1548323	1548848	525	FTL_1619	34	+	-	hypothetical protein
18	29.47	1066766	1066409	1066939	530	FTL_1120	-915	+	-	hypothetical protein
18	29.47	1066766	1066409	1066939	530	FTL_1121	6	+	-	hypothetical protein
18	29.47	1066766	1066409	1066939	530	FTL_1122	809	+	-	hypothetical protein
19	27.64	1563158	1562937	1563484	547	FTL_1632	783	-	-	hypothetical protein
19	27.64	1563158	1562937	1563484	547	FTL_1633	-9	-	-	hypothetical protein
20	27.07	1826206	1825260	1826496	1236	FTL_1893	-954	+	-	hypothetical protein, pseudogene
20	27.07	1826206	1825260	1826496	1236	FTL_1894	-832	+	-	transposase
20	27.07	1826206	1825260	1826496	1236	FTL_1895	-273	-	-	transposase, pseudogene
21	25.33	213232	213011	213476	465	FTL_0212	-506	+	-	hypothetical protein
21	25.33	213232	213011	213476	465	FTL_0213	695	+	-	hypothetical protein

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

22	24.47	553985	553819	554229	410	FTL_0575	-875	-	*isftu2*	transposase
23	23.34	315976	315828	316279	451	FTL_0332	-424	+	-	TolR protein
23	23.34	315976	315828	316279	451	FTL_0333	3	+	-	hypothetical protein
23	23.34	315976	315828	316279	451	FTL_0334	926	+	-	TolB protein precursor
24	22.59	1747840	1747696	1748136	440	FTL_1812	469	-	*hemE*	uroporphyrinogen decarboxylase
24	22.59	1747840	1747696	1748136	440	FTL_1813	251	-	-	uroporphyrinogen decarboxylase, pseudogene
25	21.5	912400	912050	912940	890	FTL_0942	-594	+	-	nicotinamide mononucleotide transport (NMT) family protein
25	21.5	912400	912050	912940	890	FTL_0943	157	+	-	Sodium/hydrogen exchanger family protein
26	19.59	860408	860116	860602	486	FTL_0881	270	-	-	hypothetical protein
27	17.98	1210447	1210126	1210629	503	FTL_1267	-622	+	-	hypothetical protein
27	17.98	1210447	1210126	1210629	503	FTL_1268	26	+	-	hypothetical protein, pseudogene
27	17.98	1210447	1210126	1210629	503	FTL_1269	129	+	-	hypothetical protein, pseudogene
27	17.98	1210447	1210126	1210629	503	FTL_1270	448	+	-	hypothetical protein, pseudogene
28	17.56	1481889	1481664	1482118	454	FTL_1550	305	-	-	lipoprotein
28	17.56	1481889	1481664	1482118	454	FTL_1551	363	+	-	hypothetical protein
29	17.21	793292	792789	793631	842	FTL_0806	155	+	-	amino acid transporter family protein
30	16.45	1265990	1265744	1266279	535	FTL_1328	460	-	-	outer membrane associated protein
30	16.45	1265990	1265744	1266279	535	FTL_1329	105	-	-	ferredoxin
30	16.45	1265990	1265744	1266279	535	FTL_1330	-388	-	*coaD*	phosphopantetheine adenylyltransferase
30	16.45	1265990	1265744	1266279	535	FTL_1331	-951	-	-	hypothetical protein
31	16.26	11476	11147	11612	465	FTL_0013	-621	+	-	regulatory protein recX
31	16.26	11476	11147	11612	465	FTL_0014	4	+	-	single-strand binding protein
31	16.26	11476	11147	11612	465	FTL_0015	586	+	-	propionate kinase
32	15.4	893020	892716	893170	454	FTL_0917	-33	-	-	transcription-repair coupling

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

										factor
32	15.4	893020	892716	893170	454	FTL_0918	-764	-	-	hypothetical protein
33	14.12	1165259	1164966	1165444	478	FTL_1213	835	-	-	hypothetical protein
33	14.12	1165259	1164966	1165444	478	FTL_1214	449	-	-	hypothetical protein
33	14.12	1165259	1164966	1165444	478	FTL_1215	-375	-	-	hypothetical protein
33	14.12	1165259	1164966	1165444	478	FTL_1216	-961	-	-	hypothetical protein
34	13.89	425386	424683	425603	920	FTL_0449	-361	+	-	hypothetical protein
34	13.89	425386	424683	425603	920	FTL_0450	-798	-	*psd*	phosphatidylserine decarboxylase
34	13.89	425386	424683	425603	920	FTL_0451	897	+	-	hypothetical protein
35	13.75	148837	148655	149124	469	FTL_0141	-831	+	-	camphor resistance protein CrcB
35	13.75	148837	148655	149124	469	FTL_0142	-443	+	-	hypothetical protein
35	13.75	148837	148655	149124	469	FTL_0143	317	+	-	hypothetical protein
35	13.75	148837	148655	149124	469	FTL_0144	-864	-	-	hypothetical protein
35	13.75	148837	148655	149124	469	FTL_0145	993	+	-	ABC transporter membrane protein
36	13.29	921376	921155	921561	406	FTL_0952	-309	-	-	hypothetical protein
36	13.29	921376	921155	921561	406	FTL_0953	-971	-	-	hypothetical protein
37	12.69	422984	422760	423361	601	FTL_0447	-48	-	-	hypothetical protein
37	12.69	422984	422760	423361	601	FTL_0448	-837	-	-	hypothetical protein
38	12.69	470861	470679	471109	430	FTL_0487	-357	+	-	maltodextrin phosphorylase
39	12.65	1017856	1017544	1017963	419	FTL_1064	-968	+	-	hypothetical protein
39	12.65	1017856	1017544	1017963	419	FTL_1065	-61	+	-	ABC transporter ATP-binding protein
39	12.65	1017856	1017544	1017963	419	FTL_1066	719	+	-	fumarylacetoacetate hydrolase family protein
40	12.17	272848	272558	272983	425	FTL_0286	136	-	-	hypothetical protein
40	12.17	272848	272558	272983	425	FTL_0287	-519	-	-	hypothetical protein
40	12.17	272848	272558	272983	425	FTL_0288	607	+	-	hypothetical protein
41	12.1	1987	1775	2205	430	FTL_0002	-200	+	-	DNA polymerase III subunit beta

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

42	11.82	472084	471866	472280	414	FTL_0488	737	+	-	4-alpha-glucanotransferase
43	11.52	1849039	1848751	1849181	430	FTL_1915	167	-	-	putative acyltransferase
44	11.51	190570	190076	190953	877	FTL_0189	285	-	-	cytochrome d terminal oxidase, polypeptide subunit I
45	11.12	840198	840021	840391	370	FTL_0858	743	-	-	hypothetical protein
45	11.12	840198	840021	840391	370	FTL_0859	513	-	-	rubredoxin
45	11.12	840198	840021	840391	370	FTL_0860	-139	-	-	hypothetical protein
45	11.12	840198	840021	840391	370	FTL_0861	442	+	*isftu2*	transposase
46	10.82	1529039	1528743	1529180	437	FTL_1599	-331	-	-	hypothetical protein
47	10.64	311720	311509	311948	439	FTL_0325	59	-	-	OmpA family protein
47	10.64	311720	311509	311948	439	FTL_0326	133	+	-	FKBP-type 16 kDa peptidyl-prolyl cis-transisomerase
47	10.64	311720	311509	311948	439	FTL_0327	578	+	*ispH*	4-hydroxy-3-methylbut-2-enyl diphosphate reductase
48	10.47	838498	838321	838757	436	FTL_0857	-21	-	-	hypothetical protein
49	10.4	357096	356777	357274	497	FTL_0387	252	-	-	aspartate aminotransferase
49	10.4	357096	356777	357274	497	FTL_0388	-49	+	-	cation transporter
49	10.4	357096	356777	357274	497	FTL_0389	924	+	-	DNA repair protein recN
50	10.24	1525426	1525248	1525825	577	FTL_1596	971	-	-	peptidyl-prolyl cis-trans isomerase
51	10.03	1307682	1307506	1307903	397	FTL_1374	-821	+	-	organic solvent tolerance protein
52	9.76	130649	130367	130742	375	FTL_0127	-778	+	-	formate dehydrogenase
52	9.76	130649	130367	130742	375	FTL_0128	-4	+	*isftu1*	transposase
53	8.94	1735070	1734735	1735387	652	FTL_1801	348	-	-	F0F1 ATP synthase subunit A
53	8.94	1735070	1734735	1735387	652	FTL_1802	-136	-	-	hypothetical protein
53	8.94	1735070	1734735	1735387	652	FTL_1803	213	+	-	major facilitator transporter
54	8.65	402579	402331	402957	626	FTL_0436	-732	-	*ileS*	isoleucyl-tRNA synthetase
55	8.55	1873077	1872956	1873323	367	FTL_1944	286	+	*isftu1*	transposase
56	8.11	1199773	1199514	1199904	390	FTL_1254	-924	+	-	exodeoxyribonuclease III, pseudogene
56	8.11	1199773	1199514	1199904	390	FTL_1255	-671	+	-	exodeoxyribonuclease III,

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

										pseudogene
56	8.11	1199773	1199514	1199904	390	FTL_1256	-55	+	-	exodeoxyribonuclease III, pseudogene
56	8.11	1199773	1199514	1199904	390	FTL_1257	585	+	-	exodeoxyribonuclease III, pseudogene
57	8	992340	991850	992475	625	FTL_1034	-34	+	-	sulfate adenylyltransferase
57	8	992340	991850	992475	625	FTL_1035	916	+	-	sulfate adenylyltransferase
58	7.99	1171072	1170805	1171390	585	FTL_1219	2	-	-	hypothetical protein
58	7.99	1171072	1170805	1171390	585	FTL_1220	227	+	-	amino-acid permease
59	7.91	780449	780213	780619	406	FTL_0794	-800	+	-	hypothetical protein, pseudogene
60	7.85	257676	257518	257880	362	FTL_0270	-850	+	*isftu2*	transposase
60	7.85	257676	257518	257880	362	FTL_0271	110	+	-	hypothetical protein
60	7.85	257676	257518	257880	362	FTL_0272	713	+	-	diaminopimelate decarboxylase
61	7.76	1392338	1392281	1392600	319	FTL_1467	-571	+	*isftu2*	transposase
62	7.67	173042	172905	173310	405	FTL_0167	727	-	-	DNA/RNA helicase
62	7.67	173042	172905	173310	405	FTL_0168	-9	-	-	hypothetical protein
62	7.67	173042	172905	173310	405	FTL_0169	-672	-	-	hypothetical protein
64	7.64	1507877	1507676	1508077	401	FTL_1581	36	+	-	lipoprotein
65	7.62	1388421	1388369	1388675	306	FTL_1462	873	-	*isftu1*	transposase
65	7.62	1388421	1388369	1388675	306	FTL_1463	-568	+	*isftu2*	transposase
66	7.38	1868230	1867989	1868416	427	FTL_1938	127	-	-	putative acyltransferase, pseudogene
66	7.38	1868230	1867989	1868416	427	FTL_1939	-458	-	-	outer membrane lipoprotein
67	7.19	584144	583915	584307	392	FTL_0597	-355	+	-	NAD dependent epimerase
67	7.19	584144	583915	584307	392	FTL_0598	633	+	-	membrane protein/O-antigen protein
68	7.15	1059402	1059244	1059654	410	FTL_1113	494	-	-	hypothetical protein
68	7.15	1059402	1059244	1059654	410	FTL_1114	-9	-	-	isochorismatase hydrolase family protein
68	7.15	1059402	1059244	1059654	410	FTL_1115	632	+	-	hypothetical protein

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

69	7.07	445879	445641	446043	402	FTL_0472	49	+	-	DNA polymerase III subunit alpha
70	6.9	960258	959961	960452	491	FTL_0990	61	-	-	lipoprotein
70	6.9	960258	959961	960452	491	FTL_0991	-571	-	-	lipoprotein, pseudogene
70	6.9	960258	959961	960452	491	FTL_0992	562	+	-	lipoprotein, pseudogene
71	6.87	634373	634185	634563	378	FTL_0651	186	-	-	transposase
72	6.77	724265	724037	724492	455	FTL_0734	-860	+	-	hypothetical protein, pseudogene
72	6.77	724265	724037	724492	455	FTL_0735	811	+	-	hypothetical protein, pseudogene
74	6.68	812104	811785	812422	637	FTL_0830	200	-	-	molybdopterin binding family protein, fragment
74	6.68	812104	811785	812422	637	FTL_0831	-76	+	-	cyanophycin synthetase
75	6.6	350080	349971	350347	376	FTL_0377	384	-	-	chorismate synthase
75	6.6	350080	349971	350347	376	FTL_0378	-410	-	-	hypothetical protein
75	6.6	350080	349971	350347	376	FTL_0379	-941	-	-	methionine sulfoxide reductase B
76	6.22	713841	713519	713928	409	FTL_0722	404	-	-	DedA family protein
76	6.22	713841	713519	713928	409	FTL_0723	131	-	-	hypothetical protein
76	6.22	713841	713519	713928	409	FTL_0724	-420	-	-	5-formyltetrahydrofolate cyclo-ligase
76	6.22	713841	713519	713928	409	FTL_0725	860	+	-	hypothetical protein
77	6.04	1609978	1609756	1610144	388	FTL_1674	219	+	-	hypothetical protein
78	6	464426	464173	464535	362	FTL_0482	702	-	-	pullulonase
79	5.77	1025170	1024903	1025270	367	FTL_1072	-59	+	-	1-deoxy-D-xylulose-5-phosphate synthase
80	5.77	670315	670250	670579	329	FTL_0682	-14	-	*isftu1*	transposase
80	5.77	670315	670250	670579	329	FTL_0683	212	+	-	transposase, pseudogene
82	5.69	649074	648845	649204	359	FTL_0666	-811	+	-	exodeoxyribonuclease V subunit gamma
83	5.52	687818	687531	687960	429	FTL_0699	112	-	-	ribosomal large subunit pseudouridine synthase D
83	5.52	687818	687531	687960	429	FTL_0700	-24	+	-	lipoprotein

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

83	5.52	687818	687531	687960	429	FTL_0701	908	+	-	FAD binding family protein
84	5.46	1439257	1439093	1439460	367	FTL_1506	295	-	-	short-chain dehydrogenase
84	5.46	1439257	1439093	1439460	367	FTL_1507	-37	-	-	3-oxoacyl-ACP reductase
84	5.46	1439257	1439093	1439460	367	FTL_1508	-660	-	-	3-oxoacyl-ACP reductase, pseudogene
85	5.35	495931	495657	496047	390	FTL_0511	-809	+	-	hypothetical protein
85	5.35	495931	495657	496047	390	FTL_0512	-387	-	-	BolA-like protein
85	5.35	495931	495657	496047	390	FTL_0513	-697	-	-	hypothetical protein
86	5.35	1229385	1229156	1229630	474	FTL_1287	571	-	*rlmL*	23S rRNA m(2)G2445 methyltransferase
86	5.35	1229385	1229156	1229630	474	FTL_1288	-58	-	-	fatty acid hydroxylase
86	5.35	1229385	1229156	1229630	474	FTL_R0033	190	+	*tRNA-Val2*	Val tRNA
88	5.06	1169417	1168963	1170075	1112	FTL_1218	522	-	-	hypothetical protein
89	5.04	1092906	1092788	1093157	369	FTL_1150	-738	-	-	Alpha,alpha-trehalase.
90	4.93	428326	428068	428428	360	FTL_0453	3	+	-	UDP-N-acetylglucosamine pyrophosphorylase/glucosamine-1-phosphate N-acetyltransferase
91	4.88	834021	833923	834252	329	FTL_0852	-493	+	-	3-phosphoshikimate 1-carboxyvinyltransferase
91	4.88	834021	833923	834252	329	FTL_0853	787	+	-	hypothetical protein
92	4.87	1625523	1625334	1625785	451	FTL_1693	944	-	-	hypothetical protein
92	4.87	1625523	1625334	1625785	451	FTL_1694	-454	-	-	hypothetical protein
93	4.87	131713	131479	131819	340	FTL_0129	-919	-	-	2-isopropylmalate synthase
94	4.79	1699084	1698836	1699202	366	FTL_1763	958	-	-	hypothetical protein
94	4.79	1699084	1698836	1699202	366	FTL_1764	-440	+	-	hypothetical protein, pseudogene
94	4.79	1699084	1698836	1699202	366	FTL_1765	658	+	-	cytochrome oxidase bd-II subunit II
95	4.79	264037	263828	264180	352	FTL_0278	128	+	-	hypothetical protein
95	4.79	264037	263828	264180	352	FTL_0279	724	+	-	hypothetical protein
96	4.77	342886	342608	342985	377	FTL_0368	497	-	-	Ser tRNA, pseudogene

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

96	4.77	342886	342608	342985	377	FTL_0369	13	-	-	Ser tRNA, pseudogene
96	4.77	342886	342608	342985	377	FTL_0370	-283	-	-	arsenical restistance operon repressor
96	4.77	342886	342608	342985	377	FTL_0371	492	+	-	hypothetical protein
97	4.67	1102502	1102248	1102676	428	FTL_1161	478	-	-	hypothetical protein
98	4.62	303442	303341	303690	349	FTL_0315	-786	+	-	MutT protein
98	4.62	303442	303341	303690	349	FTL_0316	-121	+	-	arsenate reductase
98	4.62	303442	303341	303690	349	FTL_0317	-591	-	-	hypothetical protein
99	4.6	107859	107608	108058	450	FTL_0115	474	-	-	hypothetical protein
100	4.6	92853	92655	93290	635	FTL_0094	935	-	-	ClpB protein
100	4.6	92853	92655	93290	635	FTL_0095	-715	+	-	ClpB protein, pseudogene
100	4.6	92853	92655	93290	635	FTL_0096	-416	+	-	ClpB protein, pseudogene
100	4.6	92853	92655	93290	635	FTL_0097	11	+	-	hypothetical protein
101	4.51	427484	427186	427570	384	FTL_0452	-212	+	-	bifunctional nicotinamide mononucleotide adenylyltransferase/ADP-ribose pyrophosphatase
102	4.46	1489964	1489576	1490186	610	FTL_1557	469	-	-	hypothetical protein
102	4.46	1489964	1489576	1490186	610	FTL_1558	-64	-	-	hypothetical protein
102	4.46	1489964	1489576	1490186	610	FTL_1559	288	+	-	hypothetical protein, pseudogene
103	4.37	1328363	1328254	1328589	335	FTL_1397	-701	+	-	galactokinase
103	4.37	1328363	1328254	1328589	335	FTL_1398	-839	-	-	hypothetical protein
104	4.36	1137860	1137688	1138024	336	FTL_1188	741	-	-	hypothetical protein
104	4.36	1137860	1137688	1138024	336	FTL_1189	-403	-	-	membrane-bound lytic murein transglycosylase A (MLT) family protein
104	4.36	1137860	1137688	1138024	336	FTL_1190	507	+	-	heat shock protein GrpE
105	4.34	382233	382129	382453	324	FTL_0413	150	-	-	UDP-N-acetylglucosamine 1-carboxyvinyltransferase
106	4.25	81374	81154	81504	350	FTL_0086	-16	+	-	hypothetical protein
108	4.13	899829	899602	899974	372	FTL_0926	204	-	-	Ferritin-like protein

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

108	4.13	899829	899602	899974	372	FTL_0927	2	+	-	lipoyl synthase
109	4.06	1437073	1436880	1437220	340	FTL_1504	546	-	-	peroxidase/catalase
110	3.97	796550	796464	796814	350	FTL_0808	-322	+	-	bifunctional 4'-phosphopantothenoylcysteine decarboxylase,phosphopantothenoylcysteine synthetase
110	3.97	796550	796464	796814	350	FTL_0809	850	+	-	bifunctional 4'-phosphopantothenoylcysteine decarboxylase,phosphopantothenoylcysteine synthetase, pseudogene
112	3.8	1771302	1771096	1771449	353	FTL_1836	-52	+	-	hypothetical protein
113	3.78	1378780	1378592	1379029	437	FTL_1451	286	-	-	hypothetical protein
113	3.78	1378780	1378592	1379029	437	FTL_1452	-355	-	*rpmA*	50S ribosomal protein L27
113	3.78	1378780	1378592	1379029	437	FTL_1453	-700	-	*rplU*	50S ribosomal protein L21
113	3.78	1378780	1378592	1379029	437	FTL_1454	971	+	-	50S ribosomal protein L21, pseudogene
114	3.73	429695	429594	429923	329	FTL_0454	22	+	-	glucosamine--fructose-6-phosphate aminotransferase
115	3.71	1078525	1078289	1078618	329	FTL_1134	54	-	-	hypothetical protein
115	3.71	1078525	1078289	1078618	329	FTL_1135	68	+	-	hypothetical protein, pseudogene
116	3.71	1126610	1126379	1126707	328	FTL_1174	337	-	-	cystathionine beta-synthase (cystein synthase)
116	3.71	1126610	1126379	1126707	328	FTL_1175	-935	-	-	cystathionine beta-synthase (cystein synthase), pseudogene
117	3.69	1601617	1601523	1601861	338	FTL_1668	-706	-	-	lipid A transport protein ABC transporter ATP-binding protein/permease
117	3.69	1601617	1601523	1601861	338	FTL_1669	811	+	-	tRNA CCA-pyrophosphorylase
118	3.67	52084	51868	52206	338	FTL_0050	-85	-	-	hypothetical protein
118	3.67	52084	51868	52206	338	FTL_0051	-631	-	-	hypothetical protein
119	3.67	1189554	1189452	1189803	351	FTL_1241	624	-	*isftu1*	transposase
119	3.67	1189554	1189452	1189803	351	FTL_1242	367	-	-	ThiJ/Pfpl family protein

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

119	3.67	1189554	1189452	1189803	351	FTL_1243	44	-	-	hypothetical protein
119	3.67	1189554	1189452	1189803	351	FTL_1244	240	+	-	exodeoxyribonuclease III
120	3.66	129946	129778	130059	281	FTL_R0007	-500	+	-	5S ribosomal RNA
121	3.6	1305866	1305627	1306210	583	FTL_1372	183	-	-	lipoprotein
121	3.6	1305866	1305627	1306210	583	FTL_1373	290	+	-	hypothetical protein
122	3.54	1540420	1540221	1540562	341	FTL_1611	317	-	-	glycosyl transferase family protein
122	3.54	1540420	1540221	1540562	341	FTL_1612	-215	+	-	hypothetical protein
123	3.48	152615	152442	152793	351	FTL_0146	-967	+	-	ABC transporter ATP-binding protein
123	3.48	152615	152442	152793	351	FTL_0147	436	+	-	hypothetical protein
124	3.47	1577970	1577751	1578093	342	FTL_1645	948	-	-	major facilitator transporter
124	3.47	1577970	1577751	1578093	342	FTL_1646	177	-	-	hypothetical protein
124	3.47	1577970	1577751	1578093	342	FTL_1647	-43	+	-	major facilitator transporter
125	3.46	488460	488312	488600	288	FTL_0502	-776	+	-	hypothetical protein
125	3.46	488460	488312	488600	288	FTL_0503	-965	-	*isftu1*	transposase
126	3.46	926571	926482	926803	321	FTL_0957	789	-	-	Beta-lactamase class A
126	3.46	926571	926482	926803	321	FTL_0958	-65	-	-	hypothetical protein
126	3.46	926571	926482	926803	321	FTL_0959	-923	-	-	Type IV pili leader peptidase and methylase.
127	3.4	143673	143430	143872	442	FTL_0137	-389	-	-	lipopolysaccharide protein
127	3.4	143673	143430	143872	442	FTL_0138	578	+	-	ribonuclease II family protein
128	3.39	1257767	1257529	1257864	335	FTL_1320	380	-	-	hypothetical protein
128	3.39	1257767	1257529	1257864	335	FTL_1321	100	-	-	hypothetical protein
128	3.39	1257767	1257529	1257864	335	FTL_1322	-330	-	-	hypothetical protein, pseudogene
129	3.37	506362	506130	506422	292	FTL_0526	800	+	*isftu1*	transposase
130	3.33	1476370	1476180	1476543	363	FTL_1545	966	-	-	glutamine amidotransferase subunit PdxT
130	3.33	1476370	1476180	1476543	363	FTL_1546	100	-	-	pyridoxal biosynthesis lyase PdxS
131	3.33	1167837	1167575	1168096	521	FTL_1217	612	-	-	hypothetical protein

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

132	3.31	1773896	1773786	1774113	327	FTL_1838	-458	+	-	hypothetical protein
132	3.31	1773896	1773786	1774113	327	FTL_1839	-129	+	-	lipase/acyltransferase
132	3.31	1773896	1773786	1774113	327	FTL_1840	-845	-	-	hypothetical protein
134	3.24	776325	776193	776508	315	FTL_0790	-70	+	*era*	GTP-binding protein Era
134	3.24	776325	776193	776508	315	FTL_0791	990	+	-	hypothetical protein
135	3.23	672147	672032	672348	316	FTL_0684	626	-	-	hypothetical protein
135	3.23	672147	672032	672348	316	FTL_0685	-227	-	-	NH(3)-dependent NAD(+) synthetase
136	3.21	1763697	1763471	1763803	332	FTL_1827	592	-	-	NADH dehydrogenase subunit D
136	3.21	1763697	1763471	1763803	332	FTL_1828	-74	-	-	NADH dehydrogenase I
136	3.21	1763697	1763471	1763803	332	FTL_1829	-553	-	-	NADH dehydrogenase subunit B
136	3.21	1763697	1763471	1763803	332	FTL_1830	-942	-	-	NADH dehydrogenase I subunit A
137	3.2	1130304	1130219	1130550	331	FTL_1177	147	-	*trmE*	tRNA modification GTPase TrmE
137	3.2	1130304	1130219	1130550	331	FTL_1178	-104	-	-	hypothetical protein
137	3.2	1130304	1130219	1130550	331	FTL_1179	223	+	-	sigma-54 modulation protein
137	3.2	1130304	1130219	1130550	331	FTL_1180	556	+	-	PEP-dependent sugar PTS system family protein
138	3.13	695488	695085	695624	539	FTL_0705	2	-	-	hypothetical protein
138	3.13	695488	695085	695624	539	FTL_0706	206	+	-	hypothetical protein
139	3.12	433097	432993	433316	323	FTL_0456	743	-	*rpsU*	30S ribosomal protein S21
139	3.12	433097	432993	433316	323	FTL_0457	540	-	-	cold shock protein
139	3.12	433097	432993	433316	323	FTL_0458	-36	-	-	hypothetical protein
139	3.12	433097	432993	433316	323	FTL_0459	163	+	-	methionine aminopeptidase
139	3.12	433097	432993	433316	323	FTL_0460	935	+	-	hypothetical protein
140	3.08	798365	798221	798546	325	FTL_0810	-341	-	-	cation transport regulator
140	3.08	798365	798221	798546	325	FTL_0811	-505	-	-	cation transport regulator, pseudogene
140	3.08	798365	798221	798546	325	FTL_0812	-743	-	-	cation transport regulator, pseudogene

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

141	3.05	545818	545725	546060	335	FTL_0563	346	-	-	hypothetical protein, pseudogene
141	3.05	545818	545725	546060	335	FTL_0564	-456	-	-	hypothetical protein, pseudogene
141	3.05	545818	545725	546060	335	FTL_0565	-824	-	-	hypothetical protein, pseudogene
142	3.05	6728	6553	6871	318	FTL_0006	374	-	-	indolepyruvate decarboxylase
142	3.05	6728	6553	6871	318	FTL_0007	-379	-	-	hypothetical protein
142	3.05	6728	6553	6871	318	FTL_0008	453	+	-	peptidase
143	2.97	415110	414981	415243	262	FTL_0445	-97	-	-	hypothetical protein
143	2.97	415110	414981	415243	262	FTL_R0014	-830	+	*tRNA-Ser2*	Ser tRNA
143	2.97	415110	414981	415243	262	FTL_R0015	-711	+	*tRNA-Val1*	Val tRNA
143	2.97	415110	414981	415243	262	FTL_R0016	586	+	-	16S ribosomal RNA
144	2.95	995803	995577	995907	330	FTL_1038	-422	+	-	hypothetical protein
144	2.95	995803	995577	995907	330	FTL_1039	610	+	-	hypothetical protein
144	2.95	995803	995577	995907	330	FTL_R0027	451	+	-	-
145	2.95	1248273	1248037	1248379	342	FTL_1309	440	-	-	Acetyl-CoA carboxylase beta subunit
145	2.95	1248273	1248037	1248379	342	FTL_1310	-24	-	*ndk*	nucleoside diphosphate kinase
146	2.92	372276	372065	372400	335	FTL_0401	633	-	-	Sua5_yciO_yrdC family protein
146	2.92	372276	372065	372400	335	FTL_0402	-572	+	-	intracellular septation protein A family protein
146	2.92	372276	372065	372400	335	FTL_0403	32	+	-	hypothetical protein
147	2.9	1463092	1462922	1463265	343	FTL_1531	696	-	*rumA*	23S rRNA 5-methyluridine methyltransferase
147	2.9	1463092	1462922	1463265	343	FTL_1532	35	-	-	hypothetical protein
147	2.9	1463092	1462922	1463265	343	FTL_1533	-448	-	-	DNA-directed RNA polymerase subunit omega
147	2.9	1463092	1462922	1463265	343	FTL_R0041	-165	-	*tRNA-Val3*	Val tRNA
148	2.9	1403215	1402968	1403309	341	FTL_1476	735	-	*pgi*	glucose-6-phosphate isomerase
148	2.9	1403215	1402968	1403309	341	FTL_1477	95	-	-	thiamine pyrophospokinase

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

149	2.89	640351	640132	640475	343	FTL_0657	-120	+	-	hydroxyacylglutathione hydrolase
150	2.87	17603	17331	17747	416	FTL_0018	489	-	*isftu1*	transposase
150	2.87	17603	17331	17747	416	FTL_0019	131	-	-	sugar transferase
151	2.86	628144	628038	628401	363	FTL_0644	212	-	-	carbon-nitrogen hydrolase
151	2.86	628144	628038	628401	363	FTL_0645	-43	+	-	lipoprotein
152	2.85	1043519	1043421	1043730	309	FTL_1094	278	-	-	bifunctional methionine sulfoxide reductase B/A protein, pseudogene
152	2.85	1043519	1043421	1043730	309	FTL_1095	-29	-	-	bifunctional methionine sulfoxide reductase B/A protein, pseudogene
154	2.83	810238	810156	810494	338	FTL_0828	-24	-	-	Type IV pili nucleotide binding protein, ABC transporter ATP-binding protein
154	2.83	810238	810156	810494	338	FTL_0829	182	+	-	glycerophosphoryl diester phosphodiesterase
155	2.81	850758	850585	850915	330	FTL_0869	-620	+	-	hypothetical protein
155	2.81	850758	850585	850915	330	FTL_0870	-321	+	-	hypothetical protein
155	2.81	850758	850585	850915	330	FTL_0871	61	+	-	Na+/H+ antiporter
155	2.81	850758	850585	850915	330	FTL_0872	324	+	-	hypothetical protein
156	2.78	1270860	1270774	1271114	340	FTL_1334	829	-	-	L-serine dehydratase 1
156	2.78	1270860	1270774	1271114	340	FTL_1335	105	-	-	hypothetical protein
156	2.78	1270860	1270774	1271114	340	FTL_1336	-763	-	-	prephenate dehydratase
157	2.77	1007308	1007150	1007491	341	FTL_1051	57	+	-	NADH dehydrogenase
159	2.7	1709393	1709192	1709540	348	FTL_1776	301	-	-	hypothetical protein
159	2.7	1709393	1709192	1709540	348	FTL_1777	-168	-	-	hypothetical protein, pseudogene
159	2.7	1709393	1709192	1709540	348	FTL_1778	-622	-	-	hypothetical protein, pseudogene
159	2.7	1709393	1709192	1709540	348	FTL_R0049	-920	-	*tRNA-Leu3*	Leu tRNA
160	2.65	495077	494919	495190	271	FTL_0510	205	-	*isftu1*	transposase
161	2.64	309288	309061	309455	394	FTL_0322	242	-	-	lipoprotein, pseudogene

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

161	2.64	309288	309061	309455	394	FTL_0323	-821	-	-	lipoprotein, pseudogene
162	2.6	729953	729729	730040	311	FTL_0739	195	-	-	tRNA uridine 5-carboxymethylaminomethyl modification protein GidA
162	2.6	729953	729729	730040	311	FTL_0740	-52	+	-	hypothetical protein
163	2.6	1715757	1715533	1715844	311	FTL_1783	952	-	-	dihydrolipoamide succinyltransferase component of 2-oxoglutarate dehydrogenase complex
164	2.59	971551	971424	971717	293	FTL_1006	-36	+	-	hypothetical protein, pseudogene
164	2.59	971551	971424	971717	293	FTL_1007	325	+	-	hypothetical protein, pseudogene
164	2.59	971551	971424	971717	293	FTL_1008	625	+	-	hypothetical protein, pseudogene
164	2.59	971551	971424	971717	293	FTL_1009	917	+	-	aldehyde dehydrogenase
165	2.56	13933	13761	14050	289	FTL_0016	-707	+	-	phosphate acetyltransferase
168	2.51	1743807	1743743	1744069	326	FTL_1809	-326	-	*infB*	translation initiation factor IF-2
169	2.51	1575113	1574986	1575261	275	FTL_1644	-629	-	-	glycerol kinase
170	2.5	742977	742772	743053	281	FTL_0751	-235	+	-	lipoprotein
170	2.5	742977	742772	743053	281	FTL_0752	708	+	-	hypothetical protein
171	2.49	1734011	1733770	1734094	324	FTL_1798	972	-	-	F0F1 ATP synthase subunit delta
171	2.49	1734011	1733770	1734094	324	FTL_1799	482	-	-	F0F1 ATP synthase subunit B
171	2.49	1734011	1733770	1734094	324	FTL_1800	131	-	-	F0F1 ATP synthase subunit C
172	2.47	1681175	1680937	1681259	322	FTL_1744	-233	-	*rpoB*	DNA-directed RNA polymerase subunit beta
172	2.47	1681175	1680937	1681259	322	FTL_1745	-764	-	*rplL*	50S ribosomal protein L7/L12
174	2.45	1727980	1727877	1728082	205	FTL_1793	-17	-	-	hypothetical protein
174	2.45	1727980	1727877	1728082	205	FTL_1794	-665	-	*atpC*	F0F1 ATP synthase subunit epsilon
175	2.44	383608	383394	383700	306	FTL_0414	159	-	*engA*	GTP-binding protein EngA
175	2.44	383608	383394	383700	306	FTL_0415	-596	-	-	hypothetical protein
175	2.44	383608	383394	383700	306	FTL_0416	850	+	*isftu2*	transposase

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

176	2.44	1795503	1795372	1795694	322	FTL_1863	529	-	-	glutamate decarboxylase
176	2.44	1795503	1795372	1795694	322	FTL_1864	-123	-	-	hypothetical protein
177	2.38	1457138	1456917	1457215	298	FTL_1525	721	-	-	2-C-methyl-D-erythritol 4-phosphate cytidylyltransferase
177	2.38	1457138	1456917	1457215	298	FTL_1526	441	-	-	cell division protein
177	2.38	1457138	1456917	1457215	298	FTL_1527	-937	-	*eno*	phosphopyruvate hydratase
178	2.37	1486696	1486606	1486913	307	FTL_1554	-844	+	-	succinyl-CoA synthetase, alpha subunit
178	2.37	1486696	1486606	1486913	307	FTL_1555	138	+	-	hypothetical protein
178	2.37	1486696	1486606	1486913	307	FTL_1556	962	+	-	hypothetical protein
179	2.3	851624	851490	851740	250	FTL_0873	-272	-	-	hypothetical protein, pseudogene
179	2.3	851624	851490	851740	250	FTL_0874	-712	-	-	hypothetical protein, pseudogene
179	2.3	851624	851490	851740	250	FTL_0875	809	+	*ispG*	4-hydroxy-3-methylbut-2-en-1-yl diphosphate synthase
180	2.28	559360	559118	559599	481	FTL_0580	-607	+	-	hypothetical protein
180	2.28	559360	559118	559599	481	FTL_0581	-230	+	-	sugar transport protein
181	2.28	995242	995120	995426	306	FTL_1037	-479	+	-	hypothetical protein
182	2.25	561567	561381	561636	255	FTL_0582	676	-	-	hypothetical protein
182	2.25	561567	561381	561636	255	FTL_0583	-573	-	-	acetyl-CoA acetyltransferase
183	2.23	1666875	1666613	1666942	329	FTL_1734	627	-	-	integrase/recombinase
183	2.23	1666875	1666613	1666942	329	FTL_1735	114	-	*rplS*	50S ribosomal protein L19
183	2.23	1666875	1666613	1666942	329	FTL_1736	-644	-	*trmD*	tRNA (guanine-N(1)-)-methyltransferase
184	2.17	832108	831884	832185	301	FTL_0850	-103	+	-	hypothetical protein
184	2.17	832108	831884	832185	301	FTL_0851	451	+	-	RNA polymerase factor sigma-32
185	2.16	608358	608283	608486	203	FTL_0617	272	-	-	hypothetical protein
185	2.16	608358	608283	608486	203	FTL_0618	-27	+	-	hypothetical protein, pseudogene
185	2.16	608358	608283	608486	203	FTL_0619	470	+	-	hypothetical protein, pseudogene

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

186	2.13	749415	749287	749531	244	FTL_0756	-949	+	-	hypothetical protein, pseudogene
186	2.13	749415	749287	749531	244	FTL_0757	-374	+	-	hypothetical protein, pseudogene
186	2.13	749415	749287	749531	244	FTL_0758	31	+	-	hypothetical protein, pseudogene
187	2.11	68705	68587	68889	302	FTL_0071	-915	+	-	GTP-binding protein LepA
187	2.11	68705	68587	68889	302	FTL_0072	883	+	*mnmA*	tRNA-specific 2-thiouridylase MnmA
188	2.1	863122	863023	863319	296	FTL_0883	406	-	-	metal ion transporter protein
188	2.1	863122	863023	863319	296	FTL_0884	-105	-	-	hypothetical protein
189	2.1	1603614	1603420	1603721	301	FTL_1670	-64	+	-	disulfide bond formation protein
189	2.1	1603614	1603420	1603721	301	FTL_1671	440	+	-	RND efflux transporter
190	2.05	146153	145976	146269	293	FTL_0139	285	+	-	carboxylesterase/phospholipase family protein
190	2.05	146153	145976	146269	293	FTL_0140	950	+	*hemC*	porphobilinogen deaminase
191	2.02	905158	904961	905244	283	FTL_0932	-996	+	*ruvA*	Holliday junction DNA helicase RuvA
191	2.02	905158	904961	905244	283	FTL_0933	-320	+	-	DNA recombination protein RmuC family protein
192	2	606564	606382	606705	323	FTL_0616	-490	+	-	DNA-directed RNA polymerase subunit alpha
192	2	606564	606382	606705	323	FTL_R0022	472	+	*tRNA-Leu1*	Leu tRNA
193	1.99	845804	845592	845878	286	FTL_0865	-515	+	-	major facilitator transporter
194	1.99	1201915	1201825	1201979	154	FTL_1258	-647	+	-	aldo/keto reductase
194	1.99	1201915	1201825	1201979	154	FTL_1259	320	+	-	aldo/keto reductase, pseudogene
194	1.99	1201915	1201825	1201979	154	FTL_1260	879	+	-	aldo/keto reductase, pseudogene
195	1.98	1441987	1441886	1442178	292	FTL_1510	-23	-	-	glycerol-3-phosphate transporter
196	1.98	1581635	1581473	1581718	245	FTL_1650	670	-	-	major facilitator transporter, pseudogene
196	1.98	1581635	1581473	1581718	245	FTL_1651	41	-	-	major facilitator transporter, pseudogene

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

196	1.98	1581635	1581473	1581718	245	FTL_1652	-267	-	-	major facilitator transporter, pseudogene
196	1.98	1581635	1581473	1581718	245	FTL_1653	-615	-	-	peptide transport system substrate-binding protein
197	1.97	328604	328541	328704	163	FTL_0348	-342	+	-	Ser tRNA, pseudogene
197	1.97	328604	328541	328704	163	FTL_0349	294	+	-	Ser tRNA, pseudogene
197	1.97	328604	328541	328704	163	FTL_0350	815	+	-	Ser tRNA, pseudogene
197	1.97	328604	328541	328704	163	FTL_R0013	-834	+	*tRNA-Ser1*	Ser tRNA
198	1.96	1878120	1877983	1878313	330	FTL_1949	-602	-	-	hypothetical protein
198	1.96	1878120	1877983	1878313	330	FTL_1950	648	+	-	hypothetical protein
199	1.93	224674	224646	224730	84	FTL_0222	291	-	-	hypothetical protein
199	1.93	224674	224646	224730	84	FTL_0223	-333	-	-	dihydrofolate reductase type I
199	1.93	224674	224646	224730	84	FTL_0224	526	+	*rpsB*	30S ribosomal protein S2
200	1.93	188885	188755	189035	280	FTL_0188	365	-	-	cytochrome d terminal oxidase, polypeptide subunit II
201	1.93	304105	303956	304250	294	FTL_0318	-350	-	-	lipoprotein
202	1.92	952649	952565	952733	168	FTL_0981	557	-	-	DNA polymerase IV
202	1.92	952649	952565	952733	168	FTL_0982	-52	-	-	DJ-1/Pfpl family protein
202	1.92	952649	952565	952733	168	FTL_0983	163	+	-	DJ-1/Pfpl family protein, pseudogene
203	1.91	994654	994550	994896	346	FTL_1036	-777	+	-	adenylylsulfate kinase, fragment
204	1.91	778258	778208	778490	282	FTL_0792	-619	+	-	hypothetical protein, pseudogene
204	1.91	778258	778208	778490	282	FTL_0793	326	+	-	hypothetical protein, pseudogene
205	1.9	1041463	1041339	1041601	262	FTL_1093	38	+	-	bifunctional methionine sulfoxide reductase B/A protein
206	1.9	1219369	1219290	1219551	261	FTL_1278	-26	-	-	major facilitator transporter
206	1.9	1219369	1219290	1219551	261	FTL_1279	476	+	-	acetoacetate decarboxylase, fragment
207	1.89	405817	405700	406005	305	FTL_0439	635	+	-	hypothetical protein
209	1.86	962038	961976	962215	239	FTL_0993	57	-	-	HesA/MoeB/ThiF family

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

										protein
210	1.85	1893244	1893159	1893509	350	FTL_1966	-273	-	-	anthranilate synthase component I
210	1.85	1893244	1893159	1893509	350	FTL_1967	-569	-	-	trp operon repressor
210	1.85	1893244	1893159	1893509	350	FTL_R0051	-901	-	*tRNA-Asp1*	Asp tRNA
210	1.85	1893244	1893159	1893509	350	FTL_R0052	-981	-	*tRNA-Thr1*	Thr tRNA
211	1.85	748395	748246	748514	268	FTL_0755	-227	+	-	hypothetical protein
212	1.84	1448322	1448221	1448497	276	FTL_1517	794	-	-	hypothetical protein
212	1.84	1448322	1448221	1448497	276	FTL_1518	143	-	-	hypothetical protein, pseudogene
212	1.84	1448322	1448221	1448497	276	FTL_1519	-129	-	-	hypothetical protein, pseudogene
212	1.84	1448322	1448221	1448497	276	FTL_1520	-843	-	*upp*	uracil phosphoribosyltransferase
214	1.81	1773198	1773025	1773325	300	FTL_1837	-371	+	-	hypothetical protein
215	1.81	1511720	1511589	1511892	303	FTL_1584	37	+	-	PerM family protein
216	1.8	413195	413125	413285	160	FTL_0444	-906	-	*metG*	methionyl-tRNA synthetase
217	1.77	1035075	1035006	1035143	137	FTL_1084	493	-	-	hypothetical protein
217	1.77	1035075	1035006	1035143	137	FTL_1085	199	-	-	hypothetical protein
217	1.77	1035075	1035006	1035143	137	FTL_1086	168	+	-	proton-dependent oligopeptide transporter (POT) family protein
218	1.76	589528	589396	589640	244	FTL_0601	-546	+	-	sugar transamine/perosamine synthetase
218	1.76	589528	589396	589640	244	FTL_0602	533	+	-	hypothetical protein
219	1.75	1847808	1847711	1848029	318	FTL_1914	-748	+	-	hypothetical protein
220	1.73	1523602	1523484	1523715	231	FTL_1595	558	-	*ksgA*	dimethyladenosine transferase
221	1.73	1046703	1046641	1046911	270	FTL_1098	838	-	-	bifunctional methionine sulfoxide reductase B/A protein, pseudogene
221	1.73	1046703	1046641	1046911	270	FTL_1099	282	-	-	bifunctional methionine sulfoxide reductase B/A protein, pseudogene
221	1.73	1046703	1046641	1046911	270	FTL_1100	-128	-	-	hypothetical protein

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

221	1.73	1046703	1046641	1046911	270	FTL_1101	159	+	-	hypothetical protein
221	1.73	1046703	1046641	1046911	270	FTL_1102	521	+	-	hypothetical protein, pseudogene
221	1.73	1046703	1046641	1046911	270	FTL_1103	912	+	-	hypothetical protein, pseudogene
222	1.72	800045	800011	800097	86	FTL_0813	465	-	-	cation transport regulator, pseudogene
223	1.72	969053	968958	969186	228	FTL_1003	-613	+	-	DNA polymerase III subunit epsilon
223	1.72	969053	968958	969186	228	FTL_1004	97	+	-	D-alanyl-D-alanine carboxypeptidase
223	1.72	969053	968958	969186	228	FTL_1005	557	+	-	hypothetical protein
224	1.71	1519185	1519022	1519282	260	FTL_1590	201	-	-	hypothetical protein
225	1.71	1067875	1067810	1067937	127	FTL_1123	591	+	-	hypothetical protein
226	1.7	1597680	1597507	1597766	259	FTL_1665	969	-	-	pantothenate kinase
227	1.7	730703	730536	730815	279	FTL_0741	267	+	-	hypothetical protein
228	1.69	313901	313871	313954	83	FTL_0328	-645	+	-	chorismate mutase
228	1.69	313901	313871	313954	83	FTL_0329	-268	+	-	CDP-alcohol phosphatidyltransferase
228	1.69	313901	313871	313954	83	FTL_0330	-758	-	-	hypothetical protein
228	1.69	313901	313871	313954	83	FTL_0331	928	+	-	TolQ protein
229	1.69	347571	347400	347670	270	FTL_0374	310	-	-	DNA repair protein RadA
229	1.69	347571	347400	347670	270	FTL_0375	-468	-	-	hypothetical protein
230	1.69	613378	613267	613545	278	FTL_0622	946	-	*isftu2*	transposase
230	1.69	613378	613267	613545	278	FTL_0623	-628	+	-	ABC transporter ATP-binding protein
230	1.69	613378	613267	613545	278	FTL_0624	304	+	-	ABC transporter membrane protein
231	1.69	996357	996269	996553	284	FTL_1040	760	+	-	hypothetical protein
233	1.67	620609	620462	620689	227	FTL_0631	538	-	*isftu1*	transposase
233	1.67	620609	620462	620689	227	FTL_0632	-364	+	-	ArsR family transcriptional regulator
233	1.67	620609	620462	620689	227	FTL_0633	1	+	-	hypothetical protein

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

233	1.67	620609	620462	620689	227	FTL_0634	544	+	-	NADH oxidase
233	1.67	620609	620462	620689	227	FTL_0635	730	+	-	FAD-dependent pyridine nucleotide-disulfide oxidoreductase
234	1.67	206074	206000	206237	237	FTL_0206	-371	+	-	hypothetical protein
236	1.64	99555	99393	99659	266	FTL_0103	871	-	-	hypothetical protein
236	1.64	99555	99393	99659	266	FTL_0104	458	-	-	hypothetical protein
236	1.64	99555	99393	99659	266	FTL_0105	45	-	-	hypothetical protein
236	1.64	99555	99393	99659	266	FTL_0106	178	+	-	hypothetical protein, pseudogene
236	1.64	99555	99393	99659	266	FTL_0107	756	+	*isftu1*	transposase
237	1.62	80188	79985	80248	263	FTL_0083	722	-	-	tRNA-specific 2-thiouridylase MnmA, pseudogene
237	1.62	80188	79985	80248	263	FTL_0084	140	-	-	tRNA-specific 2-thiouridylase MnmA, pseudogene
237	1.62	80188	79985	80248	263	FTL_0085	-46	+	*nhaA*	pH-dependent sodium/proton antiporter
238	1.62	122337	122253	122464	211	FTL_0125	504	-	-	hypothetical protein
239	1.61	788355	788192	788469	277	FTL_0803	-350	-	-	hypothetical protein
239	1.61	788355	788192	788469	277	FTL_0804	564	+	-	hypothetical protein
239	1.61	788355	788192	788469	277	FTL_0805	868	+	-	bifunctional proline dehydrogenase/pyrroline-5-carboxylate dehydrogenase
240	1.6	1174785	1174654	1174917	263	FTL_1223	759	-	-	hypothetical protein
240	1.6	1174785	1174654	1174917	263	FTL_1224	440	-	-	thioredoxin
240	1.6	1174785	1174654	1174917	263	FTL_1225	-253	-	-	hypothetical protein
240	1.6	1174785	1174654	1174917	263	FTL_1226	-676	-	-	hypothetical protein, pseudogene
242	1.59	1263860	1263688	1263947	259	FTL_1327	171	-	-	hypothetical protein
243	1.58	916756	916652	916895	243	FTL_0947	-207	-	-	Sodium/hydrogen exchanger family protein, pseudogene
244	1.58	1610759	1610570	1610839	269	FTL_1675	-285	-	-	hypothetical protein, pseudogene
244	1.58	1610759	1610570	1610839	269	FTL_1676	-535	-	-	hypothetical protein,

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

										pseudogene
244	1.58	1610759	1610570	1610839	269	FTL_1677	-814	-	-	hypothetical protein, pseudogene
245	1.58	441143	441076	441305	229	FTL_0465	763	-	-	BNR/Asp-box repeat-containing protein
245	1.58	441143	441076	441305	229	FTL_0466	-564	+	-	soluble lytic murein transglycosylase
246	1.58	295547	295390	295675	285	FTL_0309	11	+	*aceE*	pyruvate dehydrogenase subunit E1
248	1.54	1689342	1689177	1689399	222	FTL_1753	681	-	-	ion channel protein, fragment
248	1.54	1689342	1689177	1689399	222	FTL_1754	-10	-	-	hypothetical protein
248	1.54	1689342	1689177	1689399	222	FTL_1755	-778	-	-	glycerol uptake facilitator protein
249	1.53	1056857	1056780	1056922	142	FTL_1110	-655	+	-	hypothetical protein
249	1.53	1056857	1056780	1056922	142	FTL_1111	-326	-	-	isochorismatase hydrolase family protein
249	1.53	1056857	1056780	1056922	142	FTL_1112	553	+	*isftu2*	transposase
250	1.52	1236407	1236360	1236528	168	FTL_1296	606	-	-	amino acid antiporter
250	1.52	1236407	1236360	1236528	168	FTL_1297	-524	-	-	hypothetical protein
250	1.52	1236407	1236360	1236528	168	FTL_1298	692	+	*isftu1*	transposase
251	1.51	1105706	1105520	1105763	243	FTL_1162	-322	-	-	hypothetical protein
252	1.51	737128	737074	737185	111	FTL_0746	122	+	-	hypothetical protein
252	1.51	737128	737074	737185	111	FTL_0747	919	+	-	glutamate racemase

*Gene are referred to as associated if the annotated translational start site is within I kb upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within I kb upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S17. Genes associated* with PmrA (FTL_0552) ChIP-Seq peaks, continued

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	1223.2	1495697	1493702	1497244	3542	FTL_1567	-437	-	-	major facilitator transporter
1	1223.2	1495697	1493702	1497244	3542	FTL_1568	548	+	-	LysR family transcriptional regulator
2	77.58	148922	148478	149153	675	FTL_0141	-916	+	-	camphor resistance protein CrcB
2	77.58	148922	148478	149153	675	FTL_0142	-528	+	-	hypothetical protein
2	77.58	148922	148478	149153	675	FTL_0143	232	+	-	hypothetical protein
2	77.58	148922	148478	149153	675	FTL_0144	-779	-	-	hypothetical protein
2	77.58	148922	148478	149153	675	FTL_0145	908	+	-	ABC transporter membrane protein
3	21.85	347376	347183	347650	467	FTL_0374	115	-	-	DNA repair protein RadA
3	21.85	347376	347183	347650	467	FTL_0375	-663	-	-	hypothetical protein
4	18.13	1826571	1826316	1826835	519	FTL_1895	92	-	-	transposase, pseudogene
5	16.91	780487	780275	780631	356	FTL_0794	-838	+	-	transposase, pseudogene
6	15.27	695280	694971	695512	541	FTL_0705	-206	-	-	hypothetical protein
6	15.27	695280	694971	695512	541	FTL_0706	414	+	-	hypothetical protein
7	14.73	1156609	1156456	1156864	408	FTL_1205	721	-	-	hypothetical protein
7	14.73	1156609	1156456	1156864	408	FTL_1206	12	-	-	major facilitator transporter
7	14.73	1156609	1156456	1156864	408	FTL_1207	92	+	-	MRP like protein
7	14.73	1156609	1156456	1156864	408	FTL_1208	964	+	*dcd*	deoxycytidine triphosphate deaminase
8	11.19	1183177	1182936	1183514	578	FTL_1234	-353	+	-	hypothetical protein
8	11.19	1183177	1182936	1183514	578	FTL_1235	26	+	-	hypothetical protein, pseudogene
8	11.19	1183177	1182936	1183514	578	FTL_1236	-822	-	*infA*	translation initiation factor IF-1
8	11.19	1183177	1182936	1183514	578	FTL_1237	982	+	-	hypothetical protein

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

9	9.59	678771	678591	678922	331	FTL_0690	-458	-	-	Acyl-CoA synthetase (long-chain-fatty-acid--CoA ligase)
11	8.14	59200	58989	59343	354	FTL_0058	104	-	-	aromatic amino acid HAAP transporter
11	8.14	59200	58989	59343	354	FTL_0059	-4	+	*gidB*	16S rRNA methyltransferase GidB
11	8.14	59200	58989	59343	354	FTL_0060	610	+	-	hypothetical protein
12	8.09	1032703	1032361	1032854	493	FTL_1079	704	-	-	helix-turn-helix family protein
12	8.09	1032703	1032361	1032854	493	FTL_1080	154	-	-	hypothetical protein
12	8.09	1032703	1032361	1032854	493	FTL_1081	199	+	-	hypothetical protein
12	8.09	1032703	1032361	1032854	493	FTL_1082	587	+	-	hypothetical protein
13	7.71	828589	828365	828771	406	FTL_0847	71	+	-	preprotein translocase family protein
13	7.71	828589	828365	828771	406	FTL_0848	484	+	*secD*	preprotein translocase subunit SecD
14	6.43	162344	162133	162496	363	FTL_0156	591	-	-	phosphate transport protein
14	6.43	162344	162133	162496	363	FTL_0157	-83	-	-	hypothetical protein
14	6.43	162344	162133	162496	363	FTL_0158	390	+	-	acid phosphatase
15	5.82	996328	996005	996601	596	FTL_1038	-947	+	-	hypothetical protein
15	5.82	996328	996005	996601	596	FTL_1039	85	+	-	hypothetical protein
15	5.82	996328	996005	996601	596	FTL_1040	789	+	-	hypothetical protein
15	5.82	996328	996005	996601	596	FTL_R0027	-74	+	-	-
16	5.7	187971	187799	188131	332	FTL_0187	837	-	-	cyclohexadienyl dehydratase
16	5.7	187971	187799	188131	332	FTL_0188	-549	-	-	cytochrome d terminal oxidase, polypeptide subunit II
17	5.66	1518066	1517839	1518291	452	FTL_1588	918	-	-	mechanosensitive ion channel protein
17	5.66	1518066	1517839	1518291	452	FTL_1589	349	-	*isftu2*	transposase
17	5.66	1518066	1517839	1518291	452	FTL_1590	-918	-	-	hypothetical protein
18	5.47	22699	22401	22849	448	FTL_R0001	-447	-	*ssrA*	-
19	5.43	1782790	1782599	1782880	281	FTL_1850	178	-	-	adenylosuccinate lyase
19	5.43	1782790	1782599	1782880	281	FTL_1851	-624	-	*isftu2*	transposase

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

20	5.33	1489023	1488580	1489213	633	FTL_1557	-472	-	-	hypothetical protein
21	5.26	355641	355458	355792	334	FTL_0385	-769	+	-	hypothetical protein, pseudogene
21	5.26	355641	355458	355792	334	FTL_0386	-353	+	-	hypothetical protein, pseudogene
22	5.21	1463535	1463273	1463753	480	FTL_1532	478	-	-	hypothetical protein
22	5.21	1463535	1463273	1463753	480	FTL_1533	-5	-	-	DNA-directed RNA polymerase subunit omega
22	5.21	1463535	1463273	1463753	480	FTL_1534	-663	-	-	uridine kinase
22	5.21	1463535	1463273	1463753	480	FTL_R0041	278	-	*tRNA-Val3*	Val tRNA
23	5.21	424568	424401	424687	286	FTL_0448	747	-	-	hypothetical protein
23	5.21	424568	424401	424687	286	FTL_0449	457	+	-	hypothetical protein
24	5.03	1601683	1601533	1601867	334	FTL_1668	-640	-	-	lipid A transport protein ABC transporter ATP-binding protein/permease
24	5.03	1601683	1601533	1601867	334	FTL_1669	745	+	-	tRNA CCA-pyrophosphorylase
25	4.99	1189712	1189484	1189876	392	FTL_1241	782	-	*isftu1*	transposase
25	4.99	1189712	1189484	1189876	392	FTL_1242	525	-	-	ThiJ/Pfpl family protein
25	4.99	1189712	1189484	1189876	392	FTL_1243	202	-	-	hypothetical protein
25	4.99	1189712	1189484	1189876	392	FTL_1244	82	+	-	exodeoxyribonuclease III
25	4.99	1189712	1189484	1189876	392	FTL_1245	891	+	-	short chain dehydrogenase
26	4.82	770235	770061	770435	374	FTL_0783	861	-	*isftu1*	transposase
27	4.74	616523	616375	616700	325	FTL_0626	-923	+	-	inorganic phosphate/ATP-NAD kinase
27	4.74	616523	616375	616700	325	FTL_0627	-599	-	-	threonine efflux protein
27	4.74	616523	616375	616700	325	FTL_0628	669	+	-	threonine efflux protein, pseudogene
28	4.65	1544023	1543899	1544219	320	FTL_1614	539	-	-	UDP-N-acetylmuramoylalanine--D-glutamate ligase
28	4.65	1544023	1543899	1544219	320	FTL_1615	-558	-	*mraY*	phospho-N-acetylmuramoyl-pentapeptide-transferase
29	4.58	997646	997409	997762	353	FTL_1041	-8	+	-	octaprenyl-diphosphate synthase

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

30	4.58	325232	325120	325554	434	FTL_0344	-152	+	-	hypothetical protein
30	4.58	325232	325120	325554	434	FTL_0345	111	+	-	bile acid symporter family protein
31	4.42	1061733	1061529	1061869	340	FTL_1116	-959	+	-	ATP-dependent DNA helicase
32	4.35	1274440	1274335	1274631	296	FTL_1338	-56	-	-	alanine racemase
33	4.25	134039	133921	134252	331	FTL_0131	247	-	-	branched-chain amino acid aminotransferase
34	3.98	1384385	1384118	1384481	363	FTL_1458	122	-	-	preprotein translocase subunit SecA
34	3.98	1384385	1384118	1384481	363	FTL_1459	10	+	-	hypothetical protein
35	3.93	445475	445260	445600	340	FTL_0471	-317	+	-	hypothetical protein
35	3.93	445475	445260	445600	340	FTL_0472	453	+	-	DNA polymerase III subunit alpha
36	3.86	711515	711401	711737	336	FTL_0719	-494	+	-	hypothetical protein
36	3.86	711515	711401	711737	336	FTL_0720	-181	+	-	DNA repair protein recO
37	3.79	1549096	1548859	1549226	367	FTL_1619	-503	+	-	hypothetical protein
37	3.79	1549096	1548859	1549226	367	FTL_1620	-921	-	-	proton-dependent oligopeptide transport (POT) family protein
38	3.78	1710168	1709971	1710310	339	FTL_1777	607	-	-	sensor histidine kinase, pseudogene
38	3.78	1710168	1709971	1710310	339	FTL_1778	153	-	-	sensor histidine kinase, pseudogene
38	3.78	1710168	1709971	1710310	339	FTL_1779	-507	-	*secG*	preprotein translocase subunit SecG
38	3.78	1710168	1709971	1710310	339	FTL_R0049	-145	-	*tRNA-Leu3*	Leu tRNA
39	3.74	1179786	1179646	1180014	368	FTL_1230	390	-	-	cysteine desulfurase activator complex subunit SufB
39	3.74	1179786	1179646	1180014	368	FTL_1231	-27	-	-	hypothetical protein
39	3.74	1179786	1179646	1180014	368	FTL_1232	180	+	*trkA*	potassium transporter peripheral membrane protein
40	3.68	698988	698870	699242	372	FTL_0708	-49	-	-	hypothetical protein
40	3.68	698988	698870	699242	372	FTL_0709	219	+	-	glycosyl transferases group 1 family protein
41	3.67	957150	957060	957356	296	FTL_0986	20	-	-	ribonucleotide-diphosphate reductase subunit alpha

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

42	3.63	114133	114012	114256	244	FTL_0118	814	-	-	hypothetical protein
42	3.63	114133	114012	114256	244	FTL_0119	169	-	-	hypothetical protein
43	3.61	57859	57758	58082	324	FTL_0057	-657	+	-	hypothetical protein
44	3.6	67763	67415	67878	463	FTL_0069	-945	+	-	outer membrane lipoprotein
44	3.6	67763	67415	67878	463	FTL_0070	243	-	*rpsT*	30S ribosomal protein S20
44	3.6	67763	67415	67878	463	FTL_0071	27	+	-	GTP-binding protein LepA
45	3.52	1210893	1210716	1211035	319	FTL_1268	-420	+	-	hypothetical protein, pseudogene
45	3.52	1210893	1210716	1211035	319	FTL_1269	-317	+	-	hypothetical protein, pseudogene
45	3.52	1210893	1210716	1211035	319	FTL_1270	2	+	-	hypothetical protein, pseudogene
46	3.51	1108771	1108651	1108895	244	FTL_1164	813	-	-	hypothetical protein
46	3.51	1108771	1108651	1108895	244	FTL_1165	168	-	-	hypothetical protein
47	3.49	1747408	1747273	1747586	313	FTL_1812	37	-	*hemE*	uroporphyrinogen decarboxylase
47	3.49	1747408	1747273	1747586	313	FTL_1813	-181	-	-	uroporphyrinogen decarboxylase, pseudogene
48	3.48	618334	618140	618468	328	FTL_0629	-443	+	-	threonine efflux protein, pseudogene
48	3.48	618334	618140	618468	328	FTL_0630	466	+	-	threonine efflux protein, pseudogene
49	3.47	117544	117422	117980	558	FTL_0122	-129	-	-	hypothetical protein
49	3.47	117544	117422	117980	558	FTL_0123	-628	-	-	hypothetical protein
50	3.45	1800841	1800577	1800953	376	FTL_1868	176	-	-	multidrug resistance protein, membrane located
50	3.45	1800841	1800577	1800953	376	FTL_1869	-1	+	-	Na+/H+ antiporter
51	3.41	1772737	1772595	1772964	369	FTL_1837	90	+	-	hypothetical protein
51	3.41	1772737	1772595	1772964	369	FTL_1838	701	+	-	hypothetical protein
52	3.38	1662430	1662299	1662611	312	FTL_1730	-863	+	-	hypothetical protein
52	3.38	1662430	1662299	1662611	312	FTL_1731	502	+	-	licB-like transmembrane protein
53	3.37	541879	541490	542102	612	FTL_0558	516	-	-	oxidoreductase

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

53	3.37	541879	541490	542102	612	FTL_0559	-28	-	-	hypothetical protein
53	3.37	541879	541490	542102	612	FTL_0560	-534	-	-	hypothetical protein
54	3.34	1112184	1112072	1112640	568	FTL_1168	-128	-	-	hypothetical protein
54	3.34	1112184	1112072	1112640	568	FTL_1169	-627	-	-	hypothetical protein
55	3.32	136263	136161	136431	270	FTL_0132	-563	-	-	pyruvate phosphate dikinase
56	3.3	929044	928889	929172	283	FTL_0960	146	-	-	soluble pyridine nucleotide transhydrogenase
56	3.3	929044	928889	929172	283	FTL_0961	-809	-	-	potassium channel protein
57	3.29	744144	744004	744315	311	FTL_0752	-459	+	-	hypothetical protein
58	3.29	420815	420468	420937	469	FTL_0446	565	+	-	hypothetical protein
58	3.29	420815	420468	420937	469	FTL_R0020	-348	+	-	5S ribosomal RNA
59	3.2	2137	1796	2393	597	FTL_0002	-350	+	-	DNA polymerase III subunit beta
60	3.2	81290	81146	81478	332	FTL_0086	68	+	-	hypothetical protein
61	3.16	1554563	1554312	1554686	374	FTL_1623	247	-	-	hypothetical protein
61	3.16	1554563	1554312	1554686	374	FTL_1624	25	+	-	major facilitator transporter
62	3.16	535898	535769	536029	260	FTL_0553	-891	+	-	signal peptidase I
62	3.16	535898	535769	536029	260	FTL_0554	-21	+	*rnc*	ribonuclease III
62	3.16	535898	535769	536029	260	FTL_0555	658	+	*truB*	tRNA pseudouridine synthase B
63	3.14	746662	746565	747077	512	FTL_0753	238	-	-	aminoacylase
63	3.14	746662	746565	747077	512	FTL_0754	137	+	*xseA*	exodeoxyribonuclease VII large subunit
64	3.14	1882454	1882249	1882647	398	FTL_1954	-313	+	-	hypothetical protein
64	3.14	1882454	1882249	1882647	398	FTL_1955	-736	-	-	hypothetical protein, pseudogene
65	3.13	1077189	1077059	1077362	303	FTL_1133	16	-	-	hypothetical protein
66	3.13	1441714	1441568	1441812	244	FTL_1510	-296	-	-	glycerol-3-phosphate transporter
67	3.11	1365798	1365667	1366025	358	FTL_1434	245	-	-	DoxD-like family protein
67	3.11	1365798	1365667	1366025	358	FTL_1435	-188	-	-	hypothetical protein
67	3.11	1365798	1365667	1366025	358	FTL_1436	-463	-	-	hypothetical protein

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

68	3.08	811034	810816	811147	331	FTL_0828	772	-	-	Type IV pili nucleotide binding protein, ABC transporter ATP-binding protein
68	3.08	811034	810816	811147	331	FTL_0829	-614	+	-	glycerophosphoryl diester phosphodiesterase
68	3.08	811034	810816	811147	331	FTL_0830	-870	-	-	molybdopterin binding family protein, fragment
68	3.08	811034	810816	811147	331	FTL_0831	994	+	-	cyanophycin synthetase
69	3.06	504644	504478	504774	296	FTL_0525	519	+	-	fumerate hydratase
70	3.03	725447	725289	725537	248	FTL_0735	-371	+	-	DNA repair protein recO, pseudogene
70	3.03	725447	725289	725537	248	FTL_0736	-638	-	-	ribose-5-phosphate isomerase A
71	3.01	611362	611216	611481	265	FTL_0621	-671	+	-	sugar transamine/perosamine synthetase, pseudogene
72	3	391339	391046	391471	425	FTL_0424	-876	+	-	lipoprotein
72	3	391339	391046	391471	425	FTL_0425	-246	+	-	Type IV pili glycosylation protein
73	2.93	411232	411104	411349	245	FTL_0442	-994	+	-	hypothetical protein
73	2.93	411232	411104	411349	245	FTL_0443	-909	-	-	major facilitator transporter
75	2.9	1505915	1505697	1506022	325	FTL_1576	720	-	-	DNA mismatch repair protein
75	2.9	1505915	1505697	1506022	325	FTL_1577	377	-	-	hypothetical protein
75	2.9	1505915	1505697	1506022	325	FTL_1578	49	-	-	hypothetical protein
75	2.9	1505915	1505697	1506022	325	FTL_1579	-738	-	-	hypothetical protein
75	2.9	1505915	1505697	1506022	325	FTL_1580	892	+	-	hypothetical protein
76	2.82	944062	943888	944184	296	FTL_0974	-157	+	-	50S ribosomal protein L11 methyltransferase
76	2.82	944062	943888	944184	296	FTL_0975	678	+	-	Nif3 family protein
77	2.8	818238	818064	818342	278	FTL_0834	431	-	-	rhodanese-like family protein
77	2.8	818238	818064	818342	278	FTL_0835	-505	-	-	aromatic amino acid HAAP transporter
77	2.8	818238	818064	818342	278	FTL_0836	-788	-	-	hypothetical protein
78	2.79	761279	761105	761459	354	FTL_0769	-790	+	-	hypothetical protein
78	2.79	761279	761105	761459	354	FTL_0770	601	+	-	hypothetical protein,

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

										pseudogene
78	2.79	761279	761105	761459	354	FTL_0771	753	+	-	hypothetical protein, pseudogene
79	2.79	379184	379018	379297	279	FTL_0411	-660	-	-	hypothetical protein
81	2.74	1553618	1553307	1553789	482	FTL_1622	428	-	-	multidrug transporter (tetracycline resistance protein)
82	2.72	1576496	1576398	1576644	246	FTL_1644	754	-	-	glycerol kinase
82	2.72	1576496	1576398	1576644	246	FTL_1645	-526	-	-	major facilitator transporter
83	2.71	549510	549307	549646	339	FTL_0570	-569	+	-	hypothetical protein
83	2.71	549510	549307	549646	339	FTL_0571	7	+	-	hypothetical protein
83	2.71	549510	549307	549646	339	FTL_0572	672	+	-	hypothetical protein
84	2.71	664403	664258	664494	236	FTL_0677	482	+	-	hypothetical protein
85	2.68	1842509	1842268	1842639	371	FTL_1908	161	-	-	cell division protein FtsA
85	2.68	1842509	1842268	1842639	371	FTL_1909	-624	-	-	cell division protein FtsQ
86	2.65	1010770	1010562	1010900	338	FTL_1054	930	-	-	putative alpha-xylosidase
86	2.65	1010770	1010562	1010900	338	FTL_1055	36	-	-	putative alpha-xylosidase, pseudogene
86	2.65	1010770	1010562	1010900	338	FTL_1056	-491	-	-	putative alpha-xylosidase, pseudogene
87	2.64	338784	338624	338903	279	FTL_0362	726	-	-	Type IV pili fiber building block protein, pseudogene
87	2.64	338784	338624	338903	279	FTL_0363	126	-	-	Type IV pili fiber building block protein, pseudogene
87	2.64	338784	338624	338903	279	FTL_0364	13	+	-	Type IV pili fiber building block protein, pseudogene
87	2.64	338784	338624	338903	279	FTL_0365	379	+	-	Type IV pili fiber building block protein, pseudogene
88	2.63	1343087	1342950	1343235	285	FTL_1413	59	-	-	guanosine-3',5'-bis(diphosphate) 3'-pyrophosphohydrolase/(p)ppGpp synthase
89	2.62	500939	500775	501033	258	FTL_0519	357	-	-	septum site-determining protein MinD
89	2.62	500939	500775	501033	258	FTL_0520	-369	-	-	septum site-determining

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

										protein MinC
89	2.62	500939	500775	501033	258	FTL_0521	-559	-	*rpmG*	50S ribosomal protein L33
89	2.62	500939	500775	501033	258	FTL_0522	-822	-	*rpmB*	50S ribosomal protein L28
90	2.62	1214142	1213973	1214261	288	FTL_1273	-343	+	-	8-amino-7-oxononanoate synthase
90	2.62	1214142	1213973	1214261	288	FTL_1274	762	+	-	biotin synthesis protein BioC
91	2.61	139985	139803	140145	342	FTL_0133	89	-	-	ferrous iron transport protein
91	2.61	139985	139803	140145	342	FTL_0134	-485	-	-	hypothetical protein
92	2.59	1872651	1872568	1872823	255	FTL_1944	712	+	*isftu1*	transposase
93	2.57	121322	121158	121588	430	FTL_0125	-511	-	-	hypothetical protein
94	2.57	1152289	1152078	1152454	376	FTL_1202	86	-	-	hypothetical protein
96	2.49	892269	892150	892442	292	FTL_0917	-784	-	-	transcription-repair coupling factor
97	2.48	1115959	1115837	1116212	375	FTL_1171	-513	-	-	hypothetical protein
98	2.47	185625	185509	185836	327	FTL_0186	-701	-	-	hypothetical protein
99	2.46	1059841	1059750	1060071	321	FTL_1113	933	-	-	hypothetical protein
99	2.46	1059841	1059750	1060071	321	FTL_1114	430	-	-	isochorismatase hydrolase family protein
99	2.46	1059841	1059750	1060071	321	FTL_1115	193	+	-	hypothetical protein
100	2.46	400756	400634	400861	227	FTL_0435	239	-	*lspA*	lipoprotein signal peptidase
102	2.45	1248340	1248209	1248461	252	FTL_1309	507	-	-	Acetyl-CoA carboxylase beta subunit
102	2.45	1248340	1248209	1248461	252	FTL_1310	43	-	*ndk*	nucleoside diphosphate kinase
103	2.44	1354891	1354346	1355001	655	FTL_1423	679	-	-	glycosyl transferase family protein
103	2.44	1354891	1354346	1355001	655	FTL_1424	-297	-	-	glycosyl transferase family protein
104	2.44	1209787	1209673	1209981	308	FTL_1266	-898	+	-	lipase/esterase
104	2.44	1209787	1209673	1209981	308	FTL_1267	38	+	-	hypothetical protein
105	2.42	586923	586822	587094	272	FTL_0599	-920	+	-	glycosyl transferase family protein
105	2.42	586923	586822	587094	272	FTL_0600	180	+	-	asparagine synthase

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

106	2.42	335064	334875	335139	264	FTL_0358	114	-	-	hypothetical protein
106	2.42	335064	334875	335139	264	FTL_0359	-893	-	-	Type IV pili fiber building block protein
108	2.41	994880	994800	994986	186	FTL_1037	-117	+	-	hypothetical protein
110	2.33	123788	123604	123977	373	FTL_0126	-512	-	-	hypothetical protein
110	2.33	123788	123604	123977	373	FTL_R0003	887	+	-	16S ribosomal RNA
111	2.31	760255	760106	760367	261	FTL_0768	492	-	-	GTP binding translational elongation factor Tu and G family protein
112	2.3	977830	977669	977910	241	FTL_1017	530	-	-	cytidylate kinase
112	2.3	977830	977669	977910	241	FTL_1018	-509	-	-	phosphoserine aminotransferase
112	2.3	977830	977669	977910	241	FTL_1019	-838	-	-	hypothetical protein
112	2.3	977830	977669	977910	241	FTL_1020	829	+	*isftu1*	transposase
113	2.27	56222	56127	56356	229	FTL_0054	934	-	*isftu1*	transposase
113	2.27	56222	56127	56356	229	FTL_0055	359	-	-	hypothetical protein
113	2.27	56222	56127	56356	229	FTL_0056	-577	-	-	NADH dehydrogenase
114	2.26	1569533	1569306	1569630	324	FTL_1637	150	-	-	lipoprotein
114	2.26	1569533	1569306	1569630	324	FTL_1638	-23	+	-	3-demethylubiquinone-9 3-methyltransferase
114	2.26	1569533	1569306	1569630	324	FTL_1639	675	+	-	hypothetical protein
115	2.25	1311696	1311607	1311836	229	FTL_1376	895	-	-	carbohydrate kinase family protein (YjeF-related protein)
115	2.25	1311696	1311607	1311836	229	FTL_1377	-59	-	-	hypothetical protein
115	2.25	1311696	1311607	1311836	229	FTL_1378	-353	-	-	hypothetical protein, pseudogene
116	2.25	1230538	1230389	1230754	365	FTL_1289	-365	-	-	hypothetical protein
116	2.25	1230538	1230389	1230754	365	FTL_R0033	-963	+	*tRNA-Val2*	Val tRNA
117	2.24	1165690	1165532	1165760	228	FTL_1214	880	-	-	hypothetical protein
117	2.24	1165690	1165532	1165760	228	FTL_1215	56	-	-	hypothetical protein
117	2.24	1165690	1165532	1165760	228	FTL_1216	-530	-	-	hypothetical protein
118	2.24	1609625	1609510	1609721	211	FTL_1673	-953	+	-	major facilitator transporter

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

118	2.24	1609625	1609510	1609721	211	FTL_1674	572	+	-	hypothetical protein
119	2.23	1034175	1034004	1034272	268	FTL_1083	278	-	-	hypothetical protein
119	2.23	1034175	1034004	1034272	268	FTL_1084	-407	-	-	hypothetical protein
119	2.23	1034175	1034004	1034272	268	FTL_1085	-701	-	-	hypothetical protein
120	2.22	1769961	1769869	1770125	256	FTL_1835	13	+	-	hypothetical protein
121	2.22	800607	800419	800675	256	FTL_0814	102	-	-	hypothetical protein
121	2.22	800607	800419	800675	256	FTL_0815	-312	-	-	PRC-barrel protein
121	2.22	800607	800419	800675	256	FTL_0816	-564	-	-	hypothetical protein
122	2.22	1810016	1809959	1810195	236	FTL_1876	902	-	-	outer membrane associated protein, fragment
122	2.22	1810016	1809959	1810195	236	FTL_1877	281	-	-	outer membrane associated protein, fragment, pseudogene
123	2.19	1695679	1695592	1695970	378	FTL_1759	891	-	-	hypothetical protein
123	2.19	1695679	1695592	1695970	378	FTL_1760	360	-	-	hypothetical protein
123	2.19	1695679	1695592	1695970	378	FTL_1761	-3	-	-	hypothetical protein
123	2.19	1695679	1695592	1695970	378	FTL_1762	142	+	-	sensor histidine kinase
124	2.17	748386	748275	748526	251	FTL_0755	-218	+	-	hypothetical protein
124	2.17	748386	748275	748526	251	FTL_0756	80	+	-	hypothetical protein, pseudogene
124	2.17	748386	748275	748526	251	FTL_0757	655	+	-	hypothetical protein, pseudogene
125	2.17	1118432	1118230	1118583	353	FTL_1172	-507	-	-	hypothetical protein
125	2.17	1118432	1118230	1118583	353	FTL_R0028	882	+	-	16S ribosomal RNA
126	2.16	1243117	1243023	1243265	242	FTL_1304	531	-	-	glutamate--cysteine ligase
127	2.16	935681	935511	935752	241	FTL_0967	125	-	-	lipoate-protein ligase A
127	2.16	935681	935511	935752	241	FTL_0968	-11	+	-	tyrosyl-tRNA synthetase
128	2.15	908137	907992	908251	259	FTL_0935	938	-	-	hypothetical protein
128	2.15	908137	907992	908251	259	FTL_0936	561	-	-	hypothetical protein
128	2.15	908137	907992	908251	259	FTL_0937	-282	+	-	hypothetical protein
129	2.13	569018	568867	569209	342	FTL_0586	130	-	-	long chain fatty acid CoA ligase

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

129	2.13	569018	568867	569209	342	FTL_0587	86	+	-	arsenate reductase
129	2.13	569018	568867	569209	342	FTL_0588	482	+	-	isocitrate dehydrogenase
130	2.13	422987	422889	423059	170	FTL_0447	-45	-	-	hypothetical protein
131	2.12	947859	947739	947979	240	FTL_0977	663	+	-	hypothetical protein
132	2.12	1501689	1501426	1501815	389	FTL_1573	-554	+	-	major facilitator transporter
132	2.12	1501689	1501426	1501815	389	FTL_1574	241	+	-	hypothetical protein
133	2.12	79395	79217	79514	297	FTL_0081	897	-	-	GTP-binding protein LepA, pseudogene
133	2.12	79395	79217	79514	297	FTL_0082	303	-	-	B-type cytochrome
133	2.12	79395	79217	79514	297	FTL_0083	-71	-	-	B-type cytochrome, pseudogene
133	2.12	79395	79217	79514	297	FTL_0084	-653	-	-	B-type cytochrome, pseudogene
133	2.12	79395	79217	79514	297	FTL_0085	747	+	*nhaA*	pH-dependent sodium/proton antiporter
134	2.09	1869360	1869200	1869465	265	FTL_1939	672	-	-	outer membrane lipoprotein
134	2.09	1869360	1869200	1869465	265	FTL_1940	65	-	-	trp repressor binding protein
134	2.09	1869360	1869200	1869465	265	FTL_1941	-58	+	-	tRNA processing ribonuclease BN
135	2.06	1524813	1524701	1524962	261	FTL_1596	358	-	-	peptidyl-prolyl cis-trans isomerase
136	2.06	1636251	1636027	1636402	375	FTL_1704	216	-	-	hypothetical protein
136	2.06	1636251	1636027	1636402	375	FTL_1705	-94	+	-	cell division protein
137	2.05	11499	11274	11610	336	FTL_0013	-644	+	-	regulatory protein recX
137	2.05	11499	11274	11610	336	FTL_0014	-19	+	-	single-strand binding protein
137	2.05	11499	11274	11610	336	FTL_0015	563	+	-	propionate kinase
138	2.03	1378833	1378608	1378996	388	FTL_1451	339	-	-	hypothetical protein
138	2.03	1378833	1378608	1378996	388	FTL_1452	-302	-	*rpmA*	50S ribosomal protein L27
138	2.03	1378833	1378608	1378996	388	FTL_1453	-647	-	*rplU*	50S ribosomal protein L21
138	2.03	1378833	1378608	1378996	388	FTL_1454	918	+	-	50S ribosomal protein L21, pseudogene
139	2.03	1049497	1049410	1049726	316	FTL_1105	-55	-	-	hypothetical protein

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

139	2.03	1049497	1049410	1049726	316	FTL_1106	211	+	*alaS*	alanyl-tRNA synthetase
140	2.02	914491	914364	914676	312	FTL_0944	404	-	-	Sodium/hydrogen exchanger family protein, pseudogene
140	2.02	914491	914364	914676	312	FTL_0945	-38	-	-	hypothetical protein
140	2.02	914491	914364	914676	312	FTL_0946	395	+	-	major facilitator transporter
141	2.02	1811495	1811281	1811639	358	FTL_1878	-161	-	-	two component sensor protein kdpD
143	2	1039975	1039907	1040090	183	FTL_1090	957	-	-	rRNA methyltransferase
143	2	1039975	1039907	1040090	183	FTL_1091	-815	+	-	rRNA methyltransferase, pseudogene
143	2	1039975	1039907	1040090	183	FTL_1092	-399	+	-	rRNA methyltransferase, pseudogene
144	2	1308335	1308221	1308453	232	FTL_1375	570	+	-	heat shock protein 15 (HSP15)
145	2	1043510	1043404	1043601	197	FTL_1094	269	-	-	rRNA methyltransferase, pseudogene
145	2	1043510	1043404	1043601	197	FTL_1095	-38	-	-	rRNA methyltransferase, pseudogene
146	1.99	858164	858078	858311	233	FTL_0879	-24	-	-	beta-lactamase
146	1.99	858164	858078	858311	233	FTL_0880	453	+	-	hypothetical protein
147	1.99	939620	939499	939719	220	FTL_0971	201	+	-	FAD linked oxidase
148	1.98	1290265	1290118	1290398	280	FTL_1355	578	-	-	hypothetical protein, pseudogene
148	1.98	1290265	1290118	1290398	280	FTL_1356	111	-	-	hypothetical protein, pseudogene
148	1.98	1290265	1290118	1290398	280	FTL_1357	-874	-	-	UTP--glucose-1-phosphate uridylyltransferase
150	1.97	707593	707515	707739	224	FTL_0716	536	-	-	prolipoprotein diacylglyceryl transferase
151	1.95	1346521	1346412	1346593	181	FTL_1416	514	-	-	capsule biosynthesis protein capB
151	1.95	1346521	1346412	1346593	181	FTL_1417	-827	-	-	major facilitator transporter
152	1.95	1451795	1451696	1451894	198	FTL_1521	144	-	-	chitinase family 18 protein
152	1.95	1451795	1451696	1451894	198	FTL_1522	390	+	-	2-amino-3-ketobutyrate coenzyme A ligase
153	1.95	789164	789047	789305	258	FTL_0803	459	-	-	hypothetical protein

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

153	1.95	789164	789047	789305	258	FTL_0804	-245	+	-	hypothetical protein
153	1.95	789164	789047	789305	258	FTL_0805	59	+	-	bifunctional proline dehydrogenase/pyrroline-5-carboxylate dehydrogenase
154	1.95	757668	757528	757852	324	FTL_0767	-106	-	-	hypothetical protein
155	1.94	1212096	1211966	1212171	205	FTL_1271	-677	-	-	adenosylmethionine-8-amino-7-oxononanoate aminotransferase
155	1.94	1212096	1211966	1212171	205	FTL_1272	768	+	-	biotin synthase
156	1.93	698109	697910	698235	325	FTL_0707	168	-	-	glycosyl transferase family protein
157	1.92	38704	38479	38868	389	FTL_0036	-789	+	-	hypothetical protein
157	1.92	38704	38479	38868	389	FTL_0037	-930	-	-	hypothetical protein
158	1.92	1881085	1880800	1881223	423	FTL_1952	-469	+	-	major facilitator transporter, pseudogene
158	1.92	1881085	1880800	1881223	423	FTL_1953	-178	+	-	major facilitator transporter, pseudogene
159	1.92	896322	896178	896378	200	FTL_0920	732	-	-	hypothetical protein
159	1.92	896322	896178	896378	200	FTL_0921	97	-	-	endonuclease III
159	1.92	896322	896178	896378	200	FTL_0922	-525	-	-	Iron-sulfur cluster-binding protein
160	1.91	143970	143689	144111	422	FTL_0137	-92	-	-	lipopolysaccharide protein
160	1.91	143970	143689	144111	422	FTL_0138	281	+	-	ribonuclease II family protein
161	1.9	1877051	1876944	1877165	221	FTL_1947	294	-	-	putative ABC transporter ATP-binding protein
161	1.9	1877051	1876944	1877165	221	FTL_1948	-126	+	-	major facilitator transporter
162	1.89	154493	154368	154627	259	FTL_0148	202	+	-	Sodium/hydrogen exchanger (antiporter) family protein
163	1.87	709713	709590	709830	240	FTL_0717	110	-	-	ribonuclease E
163	1.87	709713	709590	709830	240	FTL_0718	140	+	-	cysteine desulfarase
164	1.86	1852494	1852261	1852614	353	FTL_1918	-548	+	-	30S ribosomal protein S6 modification protein-like protein
164	1.86	1852494	1852261	1852614	353	FTL_1919	471	+	-	30S ribosomal protein S6 modification protein-like

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

										protein, pseudogene
165	1.86	1174748	1174671	1174933	262	FTL_1223	722	-	-	hypothetical protein
165	1.86	1174748	1174671	1174933	262	FTL_1224	403	-	-	thioredoxin
165	1.86	1174748	1174671	1174933	262	FTL_1225	-290	-	-	hypothetical protein
165	1.86	1174748	1174671	1174933	262	FTL_1226	-713	-	-	hypothetical protein, pseudogene
166	1.85	671539	671382	671640	258	FTL_0684	18	-	-	hypothetical protein
166	1.85	671539	671382	671640	258	FTL_0685	-835	-	-	NH(3)-dependent NAD(+) synthetase
167	1.85	62424	62328	62627	299	FTL_0062	376	-	-	transcriptional regulator
167	1.85	62424	62328	62627	299	FTL_0063	132	+	-	major facilitator transporter
168	1.83	6373	6281	6427	146	FTL_0006	19	-	-	indolepyruvate decarboxylase
168	1.83	6373	6281	6427	146	FTL_0007	-734	-	-	hypothetical protein
168	1.83	6373	6281	6427	146	FTL_0008	808	+	-	peptidase
170	1.8	24760	24373	25007	634	FTL_0024	332	-	-	serine transporter
170	1.8	24760	24373	25007	634	FTL_0025	-228	-	-	hypothetical protein
170	1.8	24760	24373	25007	634	FTL_0026	720	+	-	3-hydroxyisobutyrate dehydrogenase
171	1.78	1282072	1281431	1282402	971	FTL_1347	664	-	-	hypothetical protein
171	1.78	1282072	1281431	1282402	971	FTL_1348	-371	+	-	transposase
172	1.77	1825635	1825241	1826025	784	FTL_1892	618	-	-	hypothetical protein
172	1.77	1825635	1825241	1826025	784	FTL_1893	-383	+	-	hypothetical protein, pseudogene
172	1.77	1825635	1825241	1826025	784	FTL_1894	-261	+	-	transposase
173	1.76	1126316	1126210	1126432	222	FTL_1174	43	-	-	cystathionine beta-synthase (cystein synthase)
174	1.76	1459956	1459854	1460211	357	FTL_1528	534	-	-	major facilitator transporter
174	1.76	1459956	1459854	1460211	357	FTL_1529	-333	-	-	nicotinamide mononucleotide transport (NMT) family protein
175	1.75	1661347	1661137	1661514	377	FTL_1729	219	-	-	dinucleoside polyphosphate hydrolase
176	1.74	1727970	1727900	1728044	144	FTL_1793	-27	-	-	hypothetical protein

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

176	1.74	1727970	1727900	1728044	144	FTL_1794	-675	-	*atpC*	F0F1 ATP synthase subunit epsilon
177	1.73	249317	248620	249977	1357	FTL_0264	104	-	-	cytochrome d terminal oxidase, polypeptide subunit II, pseudogene
177	1.73	249317	248620	249977	1357	FTL_0265	253	+	-	hypothetical protein
180	1.71	1256185	1255784	1256669	885	FTL_1317	625	-	-	hypothetical protein
180	1.71	1256185	1255784	1256669	885	FTL_1318	-390	+	-	hypothetical protein, pseudogene
180	1.71	1256185	1255784	1256669	885	FTL_1319	-268	+	-	transposase
181	1.71	626706	626576	626789	213	FTL_0641	591	-	-	hypothetical protein
181	1.71	626706	626576	626789	213	FTL_0642	33	-	-	lipoprotein
181	1.71	626706	626576	626789	213	FTL_0643	47	+	-	N utilisation substance protein B
182	1.71	486462	486366	486628	262	FTL_0501	-69	+	-	putative arginine decarboxylase
183	1.7	588618	588556	588748	192	FTL_0601	364	+	-	sugar transamine/perosamine synthetase
184	1.7	1425195	1425121	1425325	204	FTL_1496	580	+	-	cysteine/glutathione ABC transporter membrane/ATP-binding protein
185	1.69	912559	912492	912625	133	FTL_0942	-753	+	-	nicotinamide mononucleotide transport (NMT) family protein
185	1.69	912559	912492	912625	133	FTL_0943	-2	+	-	Sodium/hydrogen exchanger family protein
186	1.68	1184992	1184944	1185091	147	FTL_1238	3	+	-	hypothetical protein
186	1.68	1184992	1184944	1185091	147	FTL_1239	350	+	-	signal recognition particle protein, Ffh
187	1.68	1540076	1539982	1540311	329	FTL_1611	-27	-	-	glycosyl transferase family protein
187	1.68	1540076	1539982	1540311	329	FTL_1612	129	+	-	hypothetical protein
188	1.68	400129	399985	400189	204	FTL_0433	717	-	-	ribosomal large subunit methyltransferase J
188	1.68	400129	399985	400189	204	FTL_0434	-559	+	-	hypothetical protein
190	1.67	142229	142120	142387	267	FTL_0135	907	-	*isftu1*	transposase

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

190	1.67	142229	142120	142387	267	FTL_0136	-214	-	-	hypothetical protein
191	1.67	743526	743427	743596	169	FTL_0751	-784	+	-	lipoprotein
192	1.67	1200309	1200200	1200427	227	FTL_1256	-591	+	-	short chain dehydrogenase, pseudogene
192	1.67	1200309	1200200	1200427	227	FTL_1257	49	+	-	short chain dehydrogenase, pseudogene
192	1.67	1200309	1200200	1200427	227	FTL_1258	959	+	-	aldo/keto reductase
193	1.66	502340	502219	502407	188	FTL_0523	101	-	-	hypothetical protein
193	1.66	502340	502219	502407	188	FTL_0524	29	+	-	ATP-dependent DNA helicase RecG
194	1.66	1287756	1287666	1287855	189	FTL_1353	-457	+	-	hypothetical protein
195	1.65	1455579	1455489	1455795	306	FTL_1525	-838	-	-	2-C-methyl-D-erythritol 4-phosphate cytidylyltransferase
196	1.65	344313	344241	344373	132	FTL_0371	-935	+	-	hypothetical protein
196	1.65	344313	344241	344373	132	FTL_0372	38	+	*gpsA*	NAD(P)H-dependent glycerol-3-phosphate dehydrogenase
197	1.64	99669	99545	99765	220	FTL_0103	985	-	-	hypothetical protein
197	1.64	99669	99545	99765	220	FTL_0104	572	-	-	hypothetical protein
197	1.64	99669	99545	99765	220	FTL_0105	159	-	-	hypothetical protein
197	1.64	99669	99545	99765	220	FTL_0106	64	+	-	hypothetical protein, pseudogene
197	1.64	99669	99545	99765	220	FTL_0107	642	+	*isftu1*	transposase
198	1.63	343225	343116	343404	288	FTL_0368	836	-	-	Type IV pili fiber building block protein, pseudogene
198	1.63	343225	343116	343404	288	FTL_0369	352	-	-	Type IV pili fiber building block protein, pseudogene
198	1.63	343225	343116	343404	288	FTL_0370	56	-	-	arsenical restistance operon repressor
199	1.63	1493346	1493234	1493546	312	FTL_1563	-991	+	-	hypothetical protein, pseudogene
199	1.63	1493346	1493234	1493546	312	FTL_1564	453	-	-	hypothetical protein, pseudogene
199	1.63	1493346	1493234	1493546	312	FTL_1565	-43	-	-	hypothetical protein, pseudogene
199	1.63	1493346	1493234	1493546	312	FTL_1566	885	+	-	CutC family protein

Table S18. Genes associated* with FTL_1568 ChIP-Seq peaks, continued

200	1.62	1431075	1430699	1431468	769	FTL_1499	336	-	-	sulfate permease family protein
200	1.62	1431075	1430699	1431468	769	FTL_1500	-259	-	-	transposase
200	1.62	1431075	1430699	1431468	769	FTL_1501	-381	-	-	transposase, pseudogene
200	1.62	1431075	1430699	1431468	769	FTL_1502	633	+	-	major facilitator transporter
201	1.61	968496	968317	968563	246	FTL_1002	-838	+	-	hypothetical protein
201	1.61	968496	968317	968563	246	FTL_1003	-56	+	-	DNA polymerase III subunit epsilon
201	1.61	968496	968317	968563	246	FTL_1004	654	+	-	D-alanyl-D-alanine carboxypeptidase
202	1.59	837683	837586	837825	239	FTL_0856	-742	+	-	carbonic anhydrase
202	1.59	837683	837586	837825	239	FTL_0857	-836	-	-	hypothetical protein
203	1.58	172312	172221	172382	161	FTL_0167	-3	-	-	DNA/RNA helicase
203	1.58	172312	172221	172382	161	FTL_0168	-739	-	-	hypothetical protein
204	1.58	1529679	1529333	1529838	505	FTL_1599	309	-	-	hypothetical protein
205	1.57	1034770	1034692	1034901	209	FTL_1086	473	+	-	proton-dependent oligopeptide transporter (POT) family protein
206	1.55	979788	979708	979855	147	FTL_1021	-784	-	-	hypothetical protein
207	1.55	261351	261274	261507	233	FTL_0274	121	-	-	hypothetical protein, pseudogene
207	1.55	261351	261274	261507	233	FTL_0275	-647	-	-	methylated-DNA--protein-cysteine methyltransferase
207	1.55	261351	261274	261507	233	FTL_0276	677	+	-	hypothetical protein

*Gene are referred to as associated if the annotated translational start site is within I kb upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within I kb upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks

Peak Number[1]	Enrichment Factor	Max Position[2]	Start[3]	End[4]	Size	Gene Locus	Distance[5]	Strand	Gene Name	Gene Product
1	65.76	958422	957531	959135	1604	FTL_0987	129	-	-	malate dehydrogenase
1	65.76	958422	957531	959135	1604	FTL_0988	34	+	-	hypothetical protein
1	65.76	958422	957531	959135	1604	FTL_0989	562	+	-	ubiquinone biosynthesis protein
2	60.69	15176	14641	15604	963	FTL_0017	161	+	-	phosphate acetyltransferase, pseudogene
3	51.92	935393	934812	935599	787	FTL_0967	-163	-	-	lipoate-protein ligase A
3	51.92	935393	934812	935599	787	FTL_0968	277	+	-	tyrosyl-tRNA synthetase
4	50.88	1440820	1440444	1441219	775	FTL_1508	903	-	-	3-oxoacyl-ACP reductase, pseudogene
4	50.88	1440820	1440444	1441219	775	FTL_1509	142	-	-	D-alanyl-D-alanine carboxypeptidase/D-alanyl-D-alanine-endopeptidase
5	48.31	485229	484806	485709	903	FTL_0499	-558	+	-	S-adenosylmethionine decarboxylase
5	48.31	485229	484806	485709	903	FTL_0500	-70	+	-	spermidine synthase
6	46.28	767469	766819	768126	1307	FTL_0780	-695	+	-	cold shock protein (DNA-binding)
6	46.28	767469	766819	768126	1307	FTL_0781	-259	+	-	cold shock protein (DNA-binding), pseudogene
6	46.28	767469	766819	768126	1307	FTL_0782	736	+	-	cold shock protein (DNA-binding), pseudogene
7	44.76	825365	825036	825666	630	FTL_0843	-810	+	*tgt*	queuine tRNA-ribosyltransferase
9	42.21	560250	559826	560953	1127	FTL_0582	-641	-	-	hypothetical protein
10	41.85	502958	502561	503215	654	FTL_0523	719	-	-	hypothetical protein
10	41.85	502958	502561	503215	654	FTL_0524	-589	+	-	ATP-dependent DNA helicase RecG
11	41.85	232228	231846	232498	652	FTL_0233	-860	+	-	30S ribosomal protein S7
11	41.85	232228	231846	232498	652	FTL_0234	-372	+	-	elongation factor G

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

12	39.35	779857	779572	780245	673	FTL_0794	-208	+	-	aspartate aminotransferase, pseudogene
13	39.16	441510	441245	442125	880	FTL_0466	-931	+	-	soluble lytic murein transglycosylase
14	38.57	1841248	1840922	1841678	756	FTL_1907	206	-	-	cell division protein FtsZ
15	38.01	1358118	1357780	1358513	733	FTL_1427	248	-	-	hypothetical protein
16	37.15	733018	732709	733281	572	FTL_0742	674	-	-	LysR family transcriptional regulator
16	37.15	733018	732709	733281	572	FTL_0743	-117	+	-	oxidoreductase, short-chain dehydrogenase family protein
16	37.15	733018	732709	733281	572	FTL_0744	739	+	-	exodeoxyribonuclease I
17	34.9	1719738	1719165	1719940	775	FTL_1786	-448	-	-	succinate dehydrogenase, catalytic and NAD/flavoprotein subunit
17	34.9	1719738	1719165	1719940	775	FTL_1787	-829	-	-	succinate dehydrogenase hydrophobic membrane anchor protein
18	32.08	1170647	1170412	1170995	583	FTL_1219	-423	-	-	hypothetical protein
18	32.08	1170647	1170412	1170995	583	FTL_1220	652	+	-	amino-acid permease
19	31.81	12144	11741	12716	975	FTL_0014	-664	+	-	single-strand binding protein
19	31.81	12144	11741	12716	975	FTL_0015	-82	+	-	propionate kinase
20	31.14	1558963	1558552	1559636	1084	FTL_1629	-538	+	-	hypothetical protein
21	30.57	68639	68382	68846	464	FTL_0071	-849	+	-	GTP-binding protein LepA
21	30.57	68639	68382	68846	464	FTL_0072	949	+	*mnmA*	tRNA-specific 2-thiouridylase MnmA
22	30.42	17752	17558	18221	663	FTL_0018	638	-	*isftu1*	transposase
22	30.42	17752	17558	18221	663	FTL_0019	280	-	-	sugar transferase
23	30.26	883026	882767	883325	558	FTL_0906	-551	+	*engB*	ribosome biogenesis GTP-binding protein YsxC
23	30.26	883026	882767	883325	558	FTL_0907	185	+	-	ribosome biogenesis GTP-binding protein YsxC, pseudogene
23	30.26	883026	882767	883325	558	FTL_0908	579	+	-	ribosome biogenesis GTP-binding protein YsxC, pseudogene

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

24	29.56	1191686	1191274	1191987	713	FTL_1246	-373	+	-	hypothetical protein
24	29.56	1191686	1191274	1191987	713	FTL_1247	130	+	-	hypothetical protein
24	29.56	1191686	1191274	1191987	713	FTL_1248	887	+	-	glutathione reductase
25	29.55	146610	146064	147175	1111	FTL_0139	-172	+	-	carboxylesterase/phospholipase family protein
25	29.55	146610	146064	147175	1111	FTL_0140	493	+	*hemC*	porphobilinogen deaminase
26	29.46	774210	773891	774491	600	FTL_0787	-720	+	-	glycoprotease family protein
26	29.46	774210	773891	774491	600	FTL_0788	-78	+	-	glutamine amidotransferases class-II family protein
26	29.46	774210	773891	774491	600	FTL_0789	738	+	-	aspartate aminotransferase
27	29.26	245305	244965	245511	546	FTL_0257	-718	+	*rpmJ*	50S ribosomal protein L36
27	29.26	245305	244965	245511	546	FTL_0258	-488	+	*rpsM*	30S ribosomal protein S13
27	29.26	245305	244965	245511	546	FTL_0259	-93	+	-	30S ribosomal protein S11
27	29.26	245305	244965	245511	546	FTL_0260	318	+	*rpsD*	30S ribosomal protein S4
27	29.26	245305	244965	245511	546	FTL_0261	996	+	-	DNA-directed RNA polymerase subunit alpha
28	28.57	535572	535333	535804	471	FTL_0553	-565	+	-	signal peptidase I
28	28.57	535572	535333	535804	471	FTL_0554	305	+	*rnc*	ribonuclease III
28	28.57	535572	535333	535804	471	FTL_0555	984	+	*truB*	tRNA pseudouridine synthase B
29	28.23	1304894	1304576	1305135	559	FTL_1371	550	-	-	hypothetical protein
29	28.23	1304894	1304576	1305135	559	FTL_1372	-789	-	-	lipoprotein
30	26.84	1712820	1712578	1713021	443	FTL_1781	62	-	*glmM*	phosphoglucosamine mutase
30	26.84	1712820	1712578	1713021	443	FTL_1782	-476	-	-	adenine phosphoribosyltransferase
31	26.73	1578537	1578075	1578947	872	FTL_1646	744	-	-	hypothetical protein
31	26.73	1578537	1578075	1578947	872	FTL_1647	-610	+	-	major facilitator transporter
33	25.25	1326608	1326351	1326827	476	FTL_1395	65	-	-	major facilitator superfamily galactose-proton symporter
33	25.25	1326608	1326351	1326827	476	FTL_1396	26	+	-	galactose-1-phosphate uridylyltransferase
34	24.96	1246318	1246042	1246493	451	FTL_1307	594	-	-	nucleotide-binding protein, yjeE

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

34	24.96	1246318	1246042	1246493	451	FTL_1308	-598	-	-	bifunctional folylpolyglutamate synthase/ dihydrofolate synthase
35	24.92	1328590	1328192	1328753	561	FTL_1397	-928	+	-	galactokinase
35	24.92	1328590	1328192	1328753	561	FTL_1398	-612	-	-	hypothetical protein
36	24.57	97595	97358	97803	445	FTL_0100	732	-	-	carboxypeptidase,fragment
36	24.57	97595	97358	97803	445	FTL_0101	-268	-	-	voltage-gated ClC-type chloride channel clcA
36	24.57	97595	97358	97803	445	FTL_0102	-621	-	-	hypothetical protein
37	24.53	1272102	1271805	1272540	735	FTL_1336	479	-	-	prephenate dehydratase
38	24.52	488307	488047	488467	420	FTL_0502	-623	+	-	hypothetical protein
39	23.67	1010884	1010076	1011442	1366	FTL_1055	150	-	-	hypothetical protein, pseudogene
39	23.67	1010884	1010076	1011442	1366	FTL_1056	-377	-	-	hypothetical protein, pseudogene
41	23.58	633748	633545	634005	460	FTL_0650	714	-	-	prolyl-tRNA synthetase
41	23.58	633748	633545	634005	460	FTL_0651	-439	-	-	transposase
42	23.23	1305867	1305207	1306561	1354	FTL_1373	289	+	-	hypothetical protein
42	23.23	1305867	1305207	1306561	1354	FTL_1374	994	+	-	organic solvent tolerance protein
43	23.22	1808083	1807712	1808309	597	FTL_1875	19	-	-	aromatic amino acid HAAP transporter
44	23.17	527467	527281	527732	451	FTL_0544	-422	-	-	hypothetical protein
44	23.17	527467	527281	527732	451	FTL_R0021	451	-	*tRNA-Met3*	Met tRNA
45	22.84	167202	167020	167451	431	FTL_0161	856	-	-	acid phosphatase, pseudogene
45	22.84	167202	167020	167451	431	FTL_0162	112	-	-	hypothetical protein
45	22.84	167202	167020	167451	431	FTL_0163	-154	-	-	MFS transporter
46	22.71	209571	209183	209822	639	FTL_0210	-475	+	*valS*	valyl-tRNA synthetase
47	22.61	1338900	1338618	1339107	489	FTL_1409	163	-	-	glycine cleavage system protein H
47	22.61	1338900	1338618	1339107	489	FTL_1410	-59	+	*murG*	undecaprenyldiphospho-muramoylpentapeptide beta-N-

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

										acetylglucosaminyltransferase
48	22.36	847681	847431	847940	509	FTL_0866	-628	+	-	IS1 transposase
49	22.25	1685847	1685463	1686365	902	FTL_1750	938	-	*secE*	preprotein translocase subunit SecE
49	22.25	1685847	1685463	1686365	902	FTL_1751	-438	-	-	elongation factor Tu
49	22.25	1685847	1685463	1686365	902	FTL_R0045	766	-	*tRNA-Trp1*	Trp tRNA
49	22.25	1685847	1685463	1686365	902	FTL_R0046	-558	-	*tRNA-Thr2*	Thr tRNA
49	22.25	1685847	1685463	1686365	902	FTL_R0047	-660	-	*tRNA-Gly2*	Gly tRNA
49	22.25	1685847	1685463	1686365	902	FTL_R0048	-746	-	*tRNA-Tyr1*	Tyr tRNA
50	21.89	260397	260094	260587	493	FTL_0273	-827	+	-	glutamate/gamma-aminobutyrate anti-porter
50	21.89	260397	260094	260587	493	FTL_0274	-833	-	-	glutamate/gamma-aminobutyrate anti-porter, pseudogene
51	21.84	1455923	1455736	1456179	443	FTL_1525	-494	-	-	2-C-methyl-D-erythritol 4-phosphate cytidylyltransferase
51	21.84	1455923	1455736	1456179	443	FTL_1526	-774	-	-	cell division protein
52	21.58	1875485	1875284	1875781	497	FTL_1945	849	-	-	hypothetical protein
52	21.58	1875485	1875284	1875781	497	FTL_1946	361	-	-	hypothetical protein
53	20.6	1723152	1722854	1723439	585	FTL_1790	-530	+	-	major facilitator superfamily transporter
54	20.49	424590	424413	424916	503	FTL_0448	769	-	-	hypothetical protein
54	20.49	424590	424413	424916	503	FTL_0449	435	+	-	hypothetical protein
55	20.15	1529739	1529531	1530029	498	FTL_1599	369	-	-	hypothetical protein
55	20.15	1529739	1529531	1530029	498	FTL_1600	-970	-	-	periplasmic L-asparaginase II
56	20.12	839544	839289	839788	499	FTL_0858	89	-	-	hypothetical protein
56	20.12	839544	839289	839788	499	FTL_0859	-141	-	-	rubredoxin
56	20.12	839544	839289	839788	499	FTL_0860	-793	-	-	hypothetical protein
57	20.08	736110	735857	736286	429	FTL_0745	-903	+	-	excinuclease ABC subunit B
59	19.7	747593	747357	747849	492	FTL_0754	-794	+	*xseA*	exodeoxyribonuclease VII large subunit
59	19.7	747593	747357	747849	492	FTL_0755	575	+	-	hypothetical protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

59	19.7	747593	747357	747849	492	FTL_0756	873	+	-	hypothetical protein, pseudogene
60	19.48	1545636	1545440	1545885	445	FTL_1616	-606	-	-	phosphoenolpyruvate carboxykinase
60	19.48	1545636	1545440	1545885	445	FTL_1617	757	+	-	glutaminyl-tRNA synthetase
61	19.13	1651520	1651241	1651798	557	FTL_1720	-191	-	-	modification methylase
62	18.93	1350088	1349835	1350324	489	FTL_1419	171	-	-	cyanophycinase
62	18.93	1350088	1349835	1350324	489	FTL_1420	32	+	-	carbohydrate/purine kinase pfkB family protein
63	17.91	1880584	1880003	1881065	1062	FTL_1951	554	-	-	amino acid transporter LysE
63	17.91	1880584	1880003	1881065	1062	FTL_1952	32	+	-	amino acid transporter LysE, pseudogene
63	17.91	1880584	1880003	1881065	1062	FTL_1953	323	+	-	amino acid transporter LysE, pseudogene
64	17.79	876142	875991	876405	414	FTL_0897	-243	+	*miaA*	tRNA delta(2)-isopentenylpyrophosphate transferase
64	17.79	876142	875991	876405	414	FTL_0898	794	+	-	host factor I for bacteriophage Q beta replication
65	17.52	528345	528192	528737	545	FTL_0545	-378	-	-	phosphatidylcholine synthase
66	17.32	1714655	1714391	1714897	506	FTL_1783	-150	-	-	dihydrolipoamide succinyltransferase component of 2-oxoglutarate dehydrogenase complex
67	17.12	1878305	1878019	1878563	544	FTL_1949	-417	-	-	hypothetical protein
67	17.12	1878305	1878019	1878563	544	FTL_1950	463	+	-	hypothetical protein
68	16.46	945275	945081	945487	406	FTL_0975	-535	+	-	Nif3 family protein
68	16.46	945275	945081	945487	406	FTL_0976	678	+	-	hypothetical protein
69	15.94	1229706	1229527	1229963	436	FTL_1287	892	-	*rlmL*	23S rRNA m(2)G2445 methyltransferase
69	15.94	1229706	1229527	1229963	436	FTL_1288	263	-	-	fatty acid hydroxylase
69	15.94	1229706	1229527	1229963	436	FTL_R0033	-131	+	*tRNA-Val2*	Val tRNA
70	15.78	592271	592040	592494	454	FTL_0604	0	+	-	glycosyltransferase
70	15.78	592271	592040	592494	454	FTL_0605	885	+	-	glucose-1-phosphate thymidylyltransferase

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

72	15.54	161368	161085	161716	631	FTL_0155	857	-	-	transposase, pseudogene
72	15.54	161368	161085	161716	631	FTL_0156	-385	-	-	phosphate transport protein
73	15.23	1659805	1659636	1660075	439	FTL_1727	-146	-	-	N-acetyl-anhydromuranmyl-L-alanine amidase
73	15.23	1659805	1659636	1660075	439	FTL_1728	220	+	-	PAP2 family protein
74	14.89	1061311	1061146	1061577	431	FTL_1116	-537	+	-	ATP-dependent DNA helicase
75	14.77	9746	9518	9939	421	FTL_0010	-877	+	-	thiosulfate sulfurtransferase
75	14.77	9746	9518	9939	421	FTL_0011	-428	+	-	preprotein translocase subunit SecB
75	14.77	9746	9518	9939	421	FTL_0012	43	+	*recA*	recombinase A
76	14.71	1189955	1189804	1190204	400	FTL_1242	768	-	-	ThiJ/PfpI family protein
76	14.71	1189955	1189804	1190204	400	FTL_1243	445	-	-	hypothetical protein
76	14.71	1189955	1189804	1190204	400	FTL_1244	-161	+	-	exodeoxyribonuclease III
76	14.71	1189955	1189804	1190204	400	FTL_1245	648	+	-	short chain dehydrogenase
77	14.31	1276479	1276303	1276702	399	FTL_1339	508	-	-	proton-dependent oligopeptide transport (POT) family protein
77	14.31	1276479	1276303	1276702	399	FTL_1340	-324	-	-	hypothetical protein
77	14.31	1276479	1276303	1276702	399	FTL_1341	-734	-	-	hypothetical protein
77	14.31	1276479	1276303	1276702	399	FTL_1342	814	+	-	hypothetical protein
78	14.23	98535	98314	99207	893	FTL_0103	-149	-	-	hypothetical protein
78	14.23	98535	98314	99207	893	FTL_0104	-562	-	-	hypothetical protein
78	14.23	98535	98314	99207	893	FTL_0105	-975	-	-	hypothetical protein
79	13.9	514223	513755	514458	703	FTL_0532	588	-	-	hypothetical protein
79	13.9	514223	513755	514458	703	FTL_0533	-329	+	-	DNA gyrase subunit A
80	13.82	914967	914734	915130	396	FTL_0944	880	-	-	nicotinamide mononucleotide transport (NMT) family protein, pseudogene
80	13.82	914967	914734	915130	396	FTL_0945	438	-	-	hypothetical protein
80	13.82	914967	914734	915130	396	FTL_0946	-81	+	-	major facilitator transporter
81	13.75	1424947	1424750	1425147	397	FTL_1495	-950	+	-	cysteine/glutathione ABC transporter membrane/ATP-binding protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

81	13.75	1424947	1424750	1425147	397	FTL_1496	828	+	-	cysteine/glutathione ABC transporter membrane/ATP-binding protein
82	13.73	1618087	1617920	1618334	414	FTL_1684	-635	+	-	N5-glutamine S-adenosyl-L-methionine-dependent methyltransferase
82	13.73	1618087	1617920	1618334	414	FTL_1685	365	+	-	major facilitator transporter
83	13.68	1642243	1642048	1642438	390	FTL_1709	301	-	-	hypothetical protein
83	13.68	1642243	1642048	1642438	390	FTL_1710	-480	-	-	ProP osmoprotectant transporter, fragment
83	13.68	1642243	1642048	1642438	390	FTL_R0042	657	+	*tRNA-Met1*	Met tRNA
84	13.29	281309	281122	281569	447	FTL_0294	-85	-	-	DNA mismatch repair protein MutS
84	13.29	281309	281122	281569	447	FTL_0295	252	+	-	acetyl-CoA carboxylase carboxyltransferase subunit alpha
85	12.54	508917	508687	509077	390	FTL_0528	-510	+	-	Type III restriction enzyme
86	12.46	1322663	1322463	1323191	728	FTL_1392	-667	-	-	cold-shock DEAD-box protein A
86	12.46	1322663	1322463	1323191	728	FTL_1393	-980	-	-	peptidyl-prolyl cis-trans isomerase
87	12.23	505258	505070	505461	391	FTL_0525	-95	+	-	fumerate hydratase
88	12.02	1576897	1576682	1577073	391	FTL_1645	-125	-	-	major facilitator transporter
90	11.87	627285	626829	627494	665	FTL_0642	612	-	-	lipoprotein
90	11.87	627285	626829	627494	665	FTL_0643	-532	+	-	N utilisation substance protein B
90	11.87	627285	626829	627494	665	FTL_0644	-647	-	-	carbon-nitrogen hydrolase
90	11.87	627285	626829	627494	665	FTL_0645	816	+	-	lipoprotein
91	11.79	60663	60490	60901	411	FTL_0060	-853	+	-	hypothetical protein
91	11.79	60663	60490	60901	411	FTL_0061	-692	-	-	hypothetical protein
92	11.59	377841	377643	378091	448	FTL_0409	-321	-	-	purine/pyrimidine phosphoribosyl transferase family protein
92	11.59	377841	377643	378091	448	FTL_0410	299	+	-	hypothetical protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

93	11.54	454919	454508	455190	682	FTL_0477	203	+	*gcvT*	glycine cleavage system aminomethyltransferase T
94	11.45	483767	483630	484026	396	FTL_0498	-976	+	-	threonine synthase
95	11.29	1751443	1750951	1751625	674	FTL_1817	-162	-	-	NADH dehydrogenase I subunit N
96	11.28	136487	136280	136675	395	FTL_0132	-339	-	-	pyruvate phosphate dikinase
97	11.09	262623	262475	262868	393	FTL_0275	625	-	-	methylated-DNA--protein-cysteine methyltransferase
97	11.09	262623	262475	262868	393	FTL_0276	-595	+	-	hypothetical protein
97	11.09	262623	262475	262868	393	FTL_0277	127	+	-	mercuric reductase protein
98	11.07	1340759	1340566	1340946	380	FTL_1411	-793	+	-	hypothetical protein
98	11.07	1340759	1340566	1340946	380	FTL_1412	-455	+	-	recombination protein RecR
99	10.77	450463	450217	450619	402	FTL_0473	-980	+	-	peptide deformylase
99	10.77	450463	450217	450619	402	FTL_0474	318	+	-	lipoprotein releasing system, subunit C,putative membrane protein
99	10.77	450463	450217	450619	402	FTL_R0053	-284	+	-	5S ribosomal RNA
100	10.49	133257	133093	133480	387	FTL_0129	625	-	-	2-isopropylmalate synthase
100	10.49	133257	133093	133480	387	FTL_0130	363	-	-	isopropylmalate/homocitrate/citramalate synthase family protein
100	10.49	133257	133093	133480	387	FTL_0131	-535	-	-	branched-chain amino acid aminotransferase
101	10.38	541598	541335	541844	509	FTL_0558	235	-	-	oxidoreductase
101	10.38	541598	541335	541844	509	FTL_0559	-309	-	-	hypothetical protein
101	10.38	541598	541335	541844	509	FTL_0560	-815	-	-	hypothetical protein
102	10.32	179531	179321	179724	403	FTL_0177	-804	+	-	hypothetical protein
102	10.32	179531	179321	179724	403	FTL_0178	-547	+	-	inner-membrane protein
103	10.03	1076758	1076497	1076988	491	FTL_1132	724	-	-	inositol-1-monophosphatase
103	10.03	1076758	1076497	1076988	491	FTL_1133	-415	-	-	hypothetical protein
104	9.91	1294192	1293973	1294359	386	FTL_1360	169	-	*rpsU*	30S ribosomal protein S21
104	9.91	1294192	1293973	1294359	386	FTL_1361	604	+	-	cold shock protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

105	9.84	71493	71267	71667	400	FTL_0073	-186	-	-	hypothetical protein
105	9.84	71493	71267	71667	400	FTL_0074	-786	-	-	peptide deformylase
106	9.81	128575	128370	128731	361	FTL_R0007	871	+	-	5S ribosomal RNA
107	9.7	1123218	1123009	1123373	364	FTL_R0032	867	+	-	5S ribosomal RNA
110	9.61	850480	850261	850637	376	FTL_0868	809	-	*isftu2*	transposase
110	9.61	850480	850261	850637	376	FTL_0869	-342	+	-	hypothetical protein
110	9.61	850480	850261	850637	376	FTL_0870	-43	+	-	hypothetical protein
110	9.61	850480	850261	850637	376	FTL_0871	339	+	-	Na+/H+ antiporter
110	9.61	850480	850261	850637	376	FTL_0872	602	+	-	hypothetical protein
112	9.6	888305	888080	888481	401	FTL_0914	-600	+	-	hypothetical protein, pseudogene
112	9.6	888305	888080	888481	401	FTL_0915	-188	+	-	acetolactate synthase small subunit
112	9.6	888305	888080	888481	401	FTL_0916	157	+	-	ketol-acid reductoisomerase
113	9.37	419599	419392	419754	362	FTL_R0020	868	+	-	5S ribosomal RNA
116	9.18	285659	285350	285763	413	FTL_0299	333	+	*murQ*	N-acetylmuramic acid 6-phosphate etherase
117	9.01	1365430	1365179	1365577	398	FTL_1434	-123	-	-	DoxD-like family protein
117	9.01	1365430	1365179	1365577	398	FTL_1435	-556	-	-	hypothetical protein
117	9.01	1365430	1365179	1365577	398	FTL_1436	-831	-	-	hypothetical protein
118	9	511543	511339	511697	358	FTL_0529	-266	+	-	uracil-DNA glycosylase
119	8.76	1603665	1603391	1603856	465	FTL_1670	-115	+	-	disulfide bond formation protein
119	8.76	1603665	1603391	1603856	465	FTL_1671	389	+	-	RND efflux transporter
120	8.75	158155	157969	158337	368	FTL_0151	-479	+	*ipk*	4-diphosphocytidyl-2-C-methyl-D-erythritol kinase
120	8.75	158155	157969	158337	368	FTL_0152	537	+	-	hypothetical protein
120	8.75	158155	157969	158337	368	FTL_0153	881	+	*isftu1*	transposase
120	8.75	158155	157969	158337	368	FTL_R0008	367	+	*tRNA-Gln1*	Gln tRNA
121	8.65	614129	613932	614291	359	FTL_0624	-447	+	-	ABC transporter membrane protein
122	8.61	960337	960183	960548	365	FTL_0990	140	-	-	lipoprotein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

122	8.61	960337	960183	960548	365	FTL_0991	-492	-	-	lipoprotein, pseudogene
122	8.61	960337	960183	960548	365	FTL_0992	483	+	-	lipoprotein, pseudogene
124	8.51	645677	645452	645840	388	FTL_0662	694	-	-	prophage repressor protein
124	8.51	645677	645452	645840	388	FTL_0663	-525	+	-	hypothetical protein
124	8.51	645677	645452	645840	388	FTL_0664	-746	-	-	hypothetical protein
124	8.51	645677	645452	645840	388	FTL_0665	923	+	-	hypothetical protein
125	8.18	1775227	1775071	1775457	386	FTL_1840	486	-	-	hypothetical protein
125	8.18	1775227	1775071	1775457	386	FTL_1841	-938	-	*gatB*	aspartyl/glutamyl-tRNA amidotransferase subunit B
126	8.11	778685	778510	778864	354	FTL_0793	-101	+	-	aspartate aminotransferase, pseudogene
127	8.05	1042012	1041850	1042205	355	FTL_1093	-511	+	-	bifunctional methionine sulfoxide reductase B/A protein
128	8.01	236416	236249	236616	367	FTL_0238	-719	+	*rplW*	50S ribosomal protein L23
128	8.01	236416	236249	236616	367	FTL_0239	-398	+	*rplB*	50S ribosomal protein L2
128	8.01	236416	236249	236616	367	FTL_0240	441	+	*rpsS*	30S ribosomal protein S19
128	8.01	236416	236249	236616	367	FTL_0241	735	+	*rplV*	50S ribosomal protein L22
129	7.84	1536244	1536079	1536412	333	FTL_1606	957	-	-	stringent starvation protein A
129	7.84	1536244	1536079	1536412	333	FTL_1607	-868	+	-	anaerobic sulfite reductase subunit
129	7.84	1536244	1536079	1536412	333	FTL_1608	-533	+	-	hypothetical protein
129	7.84	1536244	1536079	1536412	333	FTL_1609	-178	+	-	dolichyl-phosphate-mannose-protein mannosyltransferase family protein
130	7.68	464855	464586	464958	372	FTL_0483	-825	-	-	glycogen branching protein
131	7.55	1420202	1420016	1420405	389	FTL_1491	-949	+	-	seryl-tRNA synthetase
131	7.55	1420202	1420016	1420405	389	FTL_1492	441	+	-	fructokinase
132	7.55	1580506	1580330	1580660	330	FTL_1648	405	-	-	oligopeptide transporter, subunit D, ABC transporter ATP-binding protein
132	7.55	1580506	1580330	1580660	330	FTL_1649	-105	-	-	oligopeptide transporter, subunit D, ABC transporter

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

										ATP-binding protein, pseudogene
132	7.55	1580506	1580330	1580660	330	FTL_1650	-459	-	-	oligopeptide transporter, subunit D, ABC transporter ATP-binding protein, pseudogene
133	7.54	1453584	1453390	1453721	331	FTL_1523	-180	+	*tdh*	L-threonine 3-dehydrogenase
133	7.54	1453584	1453390	1453721	331	FTL_1524	877	+	-	serine transporter
134	7.5	1403707	1403549	1403872	323	FTL_1477	587	-	-	thiamine pyrophospokinase
134	7.5	1403707	1403549	1403872	323	FTL_1478	-939	-	-	inosine-5-monophosphate dehydrogenase
135	7.5	624248	623996	624370	374	FTL_0638	-670	+	-	hypothetical protein
135	7.5	624248	623996	624370	374	FTL_0639	-680	-	-	oxidoreductase
136	7.43	1293486	1293269	1293671	402	FTL_1359	531	-	-	hypothetical protein
136	7.43	1293486	1293269	1293671	402	FTL_R0034	-387	+	*tRNA-Ser3*	Ser tRNA
137	7.42	534381	534074	534487	413	FTL_0552	-65	+	-	two-component response regulator
138	7.39	359013	358837	359261	424	FTL_0389	-993	+	-	DNA repair protein recN
139	7.36	96083	95939	96308	369	FTL_0099	812	-	-	tryptophan synthase subunit beta
140	7.35	1867052	1866905	1867580	675	FTL_1936	240	-	-	periplasmic solute binding family protein
140	7.35	1867052	1866905	1867580	675	FTL_1937	-164	-	-	periplasmic solute binding family protein, pseudogene
141	7.27	1673766	1673605	1673969	364	FTL_1742	852	-	-	hypothetical protein
142	7.14	1826475	1826298	1826662	364	FTL_1895	-4	-	-	aromatic amino acid HAAP transporter, pseudogene
143	6.99	1633182	1633014	1633382	368	FTL_1701	183	-	*glpX*	fructose 1,6-bisphosphatase II
143	6.99	1633182	1633014	1633382	368	FTL_1702	-475	-	-	hypothetical protein
144	6.94	1839747	1839573	1839938	365	FTL_1906	-127	-	*lpxC*	UDP-3-O-[3-hydroxymyristoyl] N-acetylglucosamine deacetylase
145	6.94	752326	752177	752536	359	FTL_0760	712	-	-	C32 tRNA thiolase
145	6.94	752326	752177	752536	359	FTL_0761	238	-	*smpB*	SsrA-binding protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

145	6.94	752326	752177	752536	359	FTL_0762	-163	+	-	hypothetical protein
145	6.94	752326	752177	752536	359	FTL_0763	268	+	-	hypothetical protein
145	6.94	752326	752177	752536	359	FTL_0764	615	+	-	lipoprotein
146	6.92	1083677	1083525	1083920	395	FTL_1140	646	-	-	malonyl CoA-acyl carrier protein transacylase
146	6.92	1083677	1083525	1083920	395	FTL_1141	-373	-	-	3-oxoacyl-ACP synthase
147	6.86	466176	466014	466366	352	FTL_0484	-195	+	-	phosphoglucomutase
148	6.81	1400030	1399791	1400237	446	FTL_1473	312	-	-	DNA excision repair protein subunit A
148	6.81	1400030	1399791	1400237	446	FTL_1474	-209	-	*greA*	transcription elongation factor GreA
148	6.81	1400030	1399791	1400237	446	FTL_1475	-827	-	-	Type IV pili fiber building block protein
149	6.79	1060497	1060345	1060699	354	FTL_1115	-463	+	-	hypothetical protein
151	6.74	1022826	1022591	1023018	427	FTL_1069	-978	+	-	amino acid permease
151	6.74	1022826	1022591	1023018	427	FTL_1070	320	+	-	hypothetical protein
151	6.74	1022826	1022591	1023018	427	FTL_1071	653	+	*guaA*	GMP synthase
153	6.72	142841	142594	142979	385	FTL_0136	398	-	-	hypothetical protein
154	6.7	1485935	1485705	1486099	394	FTL_1554	-83	+	-	succinyl-CoA synthetase, alpha subunit
154	6.7	1485935	1485705	1486099	394	FTL_1555	899	+	-	hypothetical protein
155	6.66	640624	640453	640792	339	FTL_0657	-393	+	-	hydroxyacylglutathione hydrolase
156	6.6	457029	456815	457151	336	FTL_0478	-774	+	-	glycine cleavage system H protein
156	6.6	457029	456815	457151	336	FTL_0479	-326	+	-	glycine dehydrogenase subunit 1
157	6.52	1551383	1551218	1551582	364	FTL_1621	-547	-	-	ABC transporter ATP-binding
159	6.34	247145	246938	247328	390	FTL_0262	172	+	*rplQ*	50S ribosomal protein L17
159	6.34	247145	246938	247328	390	FTL_0263	763	+	*isftu1*	transposase
160	6.31	919244	919137	919483	346	FTL_0948	914	-	-	hypothetical protein
160	6.31	919244	919137	919483	346	FTL_0949	-623	+	-	ribose-phosphate pyrophosphokinase

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

160	6.31	919244	919137	919483	346	FTL_0950	445	+	-	50S ribosomal protein L25
160	6.31	919244	919137	919483	346	FTL_0951	831	+	-	hypothetical protein
161	6.28	1382204	1382047	1382388	341	FTL_1456	993	-	-	transposase, pseudogene
161	6.28	1382204	1382047	1382388	341	FTL_1457	771	-	-	transposase, pseudogene
162	6.26	1110208	1110044	1110388	344	FTL_1166	162	-	-	hypothetical protein
162	6.26	1110208	1110044	1110388	344	FTL_1167	-372	-	-	hypothetical protein
163	6.18	115569	115408	115751	343	FTL_0120	162	-	-	hypothetical protein
163	6.18	115569	115408	115751	343	FTL_0121	-372	-	-	hypothetical protein
164	6.14	1444470	1444313	1444964	651	FTL_1513	-217	+	-	ABC transporter ATP-binding protein
164	6.14	1444470	1444313	1444964	651	FTL_1514	712	+	-	ABC transporter permease
165	6.07	27675	27466	27823	357	FTL_0028	383	-	-	aspartate carbamoyltransferase
166	6.05	1333437	1333196	1333542	346	FTL_1403	873	-	-	hypothetical protein
166	6.05	1333437	1333196	1333542	346	FTL_1404	406	-	*rplT*	50S ribosomal protein L20
166	6.05	1333437	1333196	1333542	346	FTL_1405	173	-	*rpmI*	50S ribosomal protein L35
166	6.05	1333437	1333196	1333542	346	FTL_1406	-337	-	-	translation initiation factor IF-3
168	5.83	911240	911052	911404	352	FTL_0939	737	-	-	aldolase/adducin class II family protein
168	5.83	911240	911052	911404	352	FTL_0940	65	-	-	methylpurine-DNA glycosylase family protein
168	5.83	911240	911052	911404	352	FTL_0941	63	+	-	hypothetical protein
168	5.83	911240	911052	911404	352	FTL_0942	566	+	-	nicotinamide mononucleotide transport (NMT) family protein
169	5.73	721812	721632	721987	355	FTL_0731	212	-	-	YhhQ family protein
169	5.73	721812	721632	721987	355	FTL_0732	-96	+	-	lactoylglutathione lyase
169	5.73	721812	721632	721987	355	FTL_0733	741	+	*isftu1*	transposase
170	5.7	955302	955121	955484	363	FTL_0984	215	-	-	bifunctional gluaredoxin/ribonucleoside-diphosphate reductase subunit beta
170	5.7	955302	955121	955484	363	FTL_0985	-52	-	-	glutaredoxin

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

171	5.63	856481	856305	856653	348	FTL_0878	-211	+	-	DNA/RNA endonuclease family protein
172	5.49	586089	585896	586403	507	FTL_0599	-86	+	-	glycosyl transferase family protein
173	5.48	1314526	1314295	1314742	447	FTL_1382	523	-	-	hypothetical protein, pseudogene
173	5.48	1314526	1314295	1314742	447	FTL_1383	11	-	-	glutathione peroxidase
173	5.48	1314526	1314295	1314742	447	FTL_1384	-365	-	-	hypothetical protein
173	5.48	1314526	1314295	1314742	447	FTL_1385	462	+	-	hypothetical protein
174	5.43	823587	823461	823770	309	FTL_0840	-778	+	-	hypothetical protein
174	5.43	823587	823461	823770	309	FTL_0841	-446	+	-	lipoprotein
174	5.43	823587	823461	823770	309	FTL_0842	-662	-	-	transposase
176	5.38	1434792	1434503	1435401	898	FTL_1503	555	-	-	deoxyguanosinetriphosphate triphosphohydrolase
177	5.35	916355	916247	916592	345	FTL_0947	-608	-	-	major facilitator transporter, pseudogene
178	5.29	355938	355799	356152	353	FTL_0386	-650	+	-	hypothetical protein, pseudogene
178	5.29	355938	355799	356152	353	FTL_0387	-906	-	-	aspartate aminotransferase
180	5.26	149202	149106	149388	282	FTL_0142	-808	+	-	hypothetical protein
180	5.26	149202	149106	149388	282	FTL_0143	-48	+	-	hypothetical protein
180	5.26	149202	149106	149388	282	FTL_0144	-499	-	-	hypothetical protein
180	5.26	149202	149106	149388	282	FTL_0145	628	+	-	ABC transporter membrane protein
182	5.17	828030	827628	828370	742	FTL_0846	-497	+	-	isochorismatase hydrolase family protein
182	5.17	828030	827628	828370	742	FTL_0847	630	+	-	preprotein translocase family protein
183	5.16	545222	545056	545474	418	FTL_0562	760	-	-	hypothetical protein, pseudogene
183	5.16	545222	545056	545474	418	FTL_0563	-250	-	-	hypothetical protein, pseudogene
184	5.14	885534	885337	885655	318	FTL_0909	-971	+	-	hypothetical protein
184	5.14	885534	885337	885655	318	FTL_0910	-636	+	-	hypothetical protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

184	5.14	885534	885337	885655	318	FTL_0911	-432	+	-	hypothetical protein, pseudogene
184	5.14	885534	885337	885655	318	FTL_0912	-203	+	-	hypothetical protein, pseudogene
184	5.14	885534	885337	885655	318	FTL_0913	984	+	-	hypothetical protein, pseudogene
185	5.04	964000	963851	964224	373	FTL_0994	525	-	-	hypothetical protein
185	5.04	964000	963851	964224	373	FTL_0995	-111	-	-	haloacid dehalogenase
185	5.04	964000	963851	964224	373	FTL_0996	207	+	-	AhpC/Tsa family protein
185	5.04	964000	963851	964224	373	FTL_0997	871	+	-	hypothetical protein
186	5.02	273733	273573	273954	381	FTL_0287	366	-	-	hypothetical protein
186	5.02	273733	273573	273954	381	FTL_0288	-278	+	-	hypothetical protein
187	4.99	1759140	1758997	1759352	355	FTL_1824	-915	-	-	NADH dehydrogenase subunit G
188	4.84	836121	835953	836296	343	FTL_0854	-748	+	-	ribonuclease H
188	4.84	836121	835953	836296	343	FTL_0855	-288	+	-	L-asparaginase
188	4.84	836121	835953	836296	343	FTL_0856	820	+	-	carbonic anhydrase
189	4.77	1451518	1451320	1451660	340	FTL_1521	-133	-	-	chitinase family 18 protein
189	4.77	1451518	1451320	1451660	340	FTL_1522	667	+	-	2-amino-3-ketobutyrate coenzyme A ligase
190	4.71	360888	360681	361008	327	FTL_0390	-167	-	-	Type IV pili fiber building block protein
190	4.71	360888	360681	361008	327	FTL_0391	-315	-	-	hypothetical protein
191	4.69	31834	31670	32003	333	FTL_0030	-55	-	-	carbamoyl phosphate synthase small subunit
191	4.69	31834	31670	32003	333	FTL_0031	472	+	-	acid phosphatase
192	4.68	1039464	1039329	1039628	299	FTL_1090	446	-	-	rRNA methyltransferase
192	4.68	1039464	1039329	1039628	299	FTL_1091	-304	+	-	rRNA methyltransferase, pseudogene
192	4.68	1039464	1039329	1039628	299	FTL_1092	112	+	-	rRNA methyltransferase, pseudogene
193	4.65	518037	517907	518249	342	FTL_0535	-368	+	-	outer membrane protein
194	4.62	367864	367684	368043	359	FTL_0397	119	+	-	phosphoribosylglycinamide formyltransferase

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

194	4.62	367864	367684	368043	359	FTL_0398	811	+	-	phosphoribosylaminoimidazole carboxylase,catalyic subunit
195	4.59	363131	362949	363254	305	FTL_0392	818	-	-	Type IV pili fiber building block protein
195	4.59	363131	362949	363254	305	FTL_0393	-434	+	-	hypothetical protein
195	4.59	363131	362949	363254	305	FTL_0394	425	+	-	bifunctional 5,10-methylene-tetrahydrofolate dehydrogenase/ 5,10-methylene-tetrahydrofolate cyclohydrolase
196	4.57	1604794	1604639	1604963	324	FTL_1672	633	+	-	AcrB/AcrD/AcrF family transporter
197	4.56	780992	780805	781123	318	FTL_0795	-582	-	*adk*	adenylate kinase
197	4.56	780992	780805	781123	318	FTL_0796	709	+	-	hypothetical protein
198	4.56	687211	687037	687421	384	FTL_0698	457	-	-	hypothetical protein
198	4.56	687211	687037	687421	384	FTL_0699	-495	-	-	ribosomal large subunit pseudouridine synthase D
198	4.56	687211	687037	687421	384	FTL_0700	583	+	-	lipoprotein
199	4.5	1711079	1710882	1711228	346	FTL_1779	404	-	*secG*	preprotein translocase subunit SecG
199	4.5	1711079	1710882	1711228	346	FTL_1780	-345	-	-	triosephosphate isomerase
199	4.5	1711079	1710882	1711228	346	FTL_R0049	766	-	*tRNA-Leu3*	Leu tRNA
200	4.49	1689857	1689556	1690030	474	FTL_1754	505	-	-	hypothetical protein
200	4.49	1689857	1689556	1690030	474	FTL_1755	-263	-	-	glycerol uptake facilitator protein
201	4.48	1136061	1135921	1136287	366	FTL_1186	709	-	*rpsI*	30S ribosomal protein S9
201	4.48	1136061	1135921	1136287	366	FTL_1187	265	-	*rplM*	50S ribosomal protein L13
203	4.42	1672666	1672491	1672839	348	FTL_1741	-12	-	-	putative DNA-binding/iron metalloprotein/AP endonuclease
204	4.36	1050299	1050169	1050471	302	FTL_1105	747	-	-	hypothetical protein
204	4.36	1050299	1050169	1050471	302	FTL_1106	-591	+	*alaS*	alanyl-tRNA synthetase
205	4.36	79704	79402	79894	492	FTL_0082	612	-	-	B-type cytochrome
205	4.36	79704	79402	79894	492	FTL_0083	238	-	-	B-type cytochrome,

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

										pseudogene
205	4.36	79704	79402	79894	492	FTL_0084	-344	-	-	B-type cytochrome, pseudogene
205	4.36	79704	79402	79894	492	FTL_0085	438	+	*nhaA*	pH-dependent sodium/proton antiporter
206	4.35	1342960	1342759	1343091	332	FTL_1413	-68	-	-	guanosine-3',5'-bis(diphosphate) 3'-pyrophosphohydrolase/(p)ppGpp synthase
207	4.32	814850	814672	814991	319	FTL_0832	26	+	-	hypothetical protein
208	4.26	469857	469699	470002	303	FTL_0486	-927	+	*glgA*	glycogen synthase
208	4.26	469857	469699	470002	303	FTL_0487	647	+	-	maltodextrin phosphorylase
209	4.18	386993	386777	387154	377	FTL_0418	807	-	-	hypothetical protein
210	4.17	1489028	1488882	1489438	556	FTL_1557	-467	-	-	hypothetical protein
210	4.17	1489028	1488882	1489438	556	FTL_1558	-1000	-	-	hypothetical protein
211	4.17	1122026	1121865	1122182	317	FTL_R0031	-872	+	-	23S ribosomal RNA
212	4.17	1457539	1457375	1457816	441	FTL_1527	-536	-	*eno*	phosphopyruvate hydratase
213	4.12	1512413	1512188	1512552	364	FTL_1584	-656	+	-	PerM family protein
213	4.12	1512413	1512188	1512552	364	FTL_1585	444	+	-	single-stranded-DNA-specific exonuclease
214	4.07	127391	127224	127542	318	FTL_R0006	-876	+	-	23S ribosomal RNA
215	4.02	458666	458252	458916	664	FTL_0480	-590	+	-	glycine dehydrogenase subunit 2
216	3.98	797103	796952	797279	327	FTL_0808	-875	+	-	bifunctional 4'-phosphopantothenoylcysteine decarboxylase,phosphopantothenoylcysteine synthetase
216	3.98	797103	796952	797279	327	FTL_0809	297	+	-	bifunctional 4'-phosphopantothenoylcysteine decarboxylase,phosphopantothenoylcysteine synthetase, pseudogene
217	3.94	112269	112097	112447	350	FTL_0116	880	-	-	hypothetical protein
217	3.94	112269	112097	112447	350	FTL_0117	67	-	-	hypothetical protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

218	3.91	88447	88276	88574	298	FTL_0093	-641	-	-	chitinase
219	3.9	1856490	1856292	1856597	305	FTL_1922	588	-	-	YggT family protein
219	3.9	1856490	1856292	1856597	305	FTL_1923	-342	-	-	hypothetical protein
219	3.9	1856490	1856292	1856597	305	FTL_1924	-807	-	-	hypothetical protein, pseudogene
220	3.89	1422659	1422406	1422767	361	FTL_1493	-988	+	-	polysaccharide biosynthesis protein (export protein)
220	3.89	1422659	1422406	1422767	361	FTL_1494	-961	-	-	hypothetical protein
221	3.88	1021151	1020863	1021330	467	FTL_1068	-135	+	*truA*	tRNA pseudouridine synthase A
222	3.87	1409480	1409312	1409648	336	FTL_1482	-977	+	-	peptidase M16 family protein
222	3.87	1409480	1409312	1409648	336	FTL_1483	283	+	-	peptidase M16 family protein
224	3.85	174038	173897	174223	326	FTL_0168	987	-	-	hypothetical protein
224	3.85	174038	173897	174223	326	FTL_0169	324	-	-	hypothetical protein
224	3.85	174038	173897	174223	326	FTL_0170	75	+	-	hypothetical protein
225	3.84	769648	769530	769880	350	FTL_0783	274	-	*isftu1*	transposase
227	3.82	1610793	1610663	1610982	319	FTL_1674	-596	+	-	hypothetical protein
227	3.82	1610793	1610663	1610982	319	FTL_1675	-251	-	-	hypothetical protein, pseudogene
227	3.82	1610793	1610663	1610982	319	FTL_1676	-501	-	-	hypothetical protein, pseudogene
227	3.82	1610793	1610663	1610982	319	FTL_1677	-780	-	-	hypothetical protein, pseudogene
228	3.79	556609	556433	556745	312	FTL_0577	-900	+	-	hypothetical protein
228	3.79	556609	556433	556745	312	FTL_0578	-275	+	-	ornithine cyclodeaminase
228	3.79	556609	556433	556745	312	FTL_0579	644	+	-	putative nicotinate phosphoribosyltransferase
229	3.77	1567022	1566882	1567183	301	FTL_1636	-55	+	-	hypothetical protein
230	3.77	1243746	1243523	1243877	354	FTL_1305	-434	-	-	virulence factor MviN
231	3.76	1106900	1106733	1107088	355	FTL_1162	872	-	-	hypothetical protein
231	3.76	1106900	1106733	1107088	355	FTL_1163	59	-	-	hypothetical protein
232	3.74	598606	598423	598726	303	FTL_0609	-288	+	-	phosphomannomutase

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

233	3.73	1245724	1245566	1245924	358	FTL_1306	460	-	-	hypothetical protein
234	3.71	943105	942951	943259	308	FTL_0973	-580	+	-	tyrosyl-tRNA synthetase, pseudogene
234	3.71	943105	942951	943259	308	FTL_0974	800	+	-	50S ribosomal protein L11 methyltransferase
235	3.7	601595	601475	601736	261	FTL_0610	-562	-	*rho*	transcription termination factor Rho
235	3.7	601595	601475	601736	261	FTL_0611	-928	-	-	thioredoxin
236	3.68	418398	418248	418564	316	FTL_R0018	-992	+	*tRNA-Ala2*	Ala tRNA
236	3.68	418398	418248	418564	316	FTL_R0019	-862	+	-	23S ribosomal RNA
237	3.66	284291	284104	284482	378	FTL_0298	-257	+	-	hypothetical protein
238	3.64	894683	894526	894843	317	FTL_0918	899	-	-	hypothetical protein
238	3.64	894683	894526	894843	317	FTL_0919	-476	-	-	hypothetical protein
238	3.64	894683	894526	894843	317	FTL_0920	-907	-	-	hypothetical protein
239	3.62	151155	151018	151327	309	FTL_0146	493	+	-	ABC transporter ATP-binding protein
240	3.62	1777817	1777682	1778003	321	FTL_1842	192	-	*gatA*	aspartyl/glutamyl-tRNA amidotransferase subunit A
240	3.62	1777817	1777682	1778003	321	FTL_1843	-98	-	-	Glu-tRNAGln amidotransferase C subunit
240	3.62	1777817	1777682	1778003	321	FTL_1844	-669	-	-	secretion protein
241	3.6	835046	834841	835143	302	FTL_0853	-238	+	-	hypothetical protein
243	3.57	584092	583907	584217	310	FTL_0597	-303	+	-	NAD dependent epimerase
243	3.57	584092	583907	584217	310	FTL_0598	685	+	-	membrane protein/O-antigen protein
245	3.56	231608	231476	231773	297	FTL_0232	-653	+	*rpsL*	30S ribosomal protein S12
247	3.5	203712	203594	203909	315	FTL_0203	-725	+	-	hypothetical protein
247	3.5	203712	203594	203909	315	FTL_0204	278	+	-	hypothetical protein
248	3.48	784306	783998	784471	473	FTL_0799	-440	+	-	Type IV pili lipoprotein
248	3.48	784306	783998	784471	473	FTL_0800	163	+	-	Type IV pilin multimeric outer membrane protein
249	3.46	579122	578908	579225	317	FTL_0592	-896	+	-	dTDP-glucose 4,6-dehydratase

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

249	3.46	579122	578908	579225	317	FTL_0593	833	+	-	galactosyl transferase
250	3.44	1344333	1344133	1344451	318	FTL_1414	25	-	-	hypothetical protein
250	3.44	1344333	1344133	1344451	318	FTL_1415	-441	-	-	capsule biosynthesis protein CapC
252	3.39	291733	291561	291866	305	FTL_0304	376	-	-	Na+/H+ antiporter
252	3.39	291733	291561	291866	305	FTL_0305	-359	-	-	hypothetical protein
253	3.37	218490	218350	218689	339	FTL_0218	-244	+	*gltX*	glutamyl-tRNA synthetase
254	3.31	267490	267367	267664	297	FTL_0282	-385	-	-	hypothetical protein
254	3.31	267490	267367	267664	297	FTL_0283	576	+	-	aromatic amino acid HAAP transporter
255	3.3	1054337	1054198	1054483	285	FTL_1108	-639	+	-	cytosol aminopeptidase
255	3.3	1054337	1054198	1054483	285	FTL_1109	874	+	-	transaldolase B
256	3.3	762922	762805	763071	266	FTL_0771	-890	+	-	hypothetical protein, pseudogene
256	3.3	762922	762805	763071	266	FTL_0772	-163	+	-	hypothetical protein, pseudogene
256	3.3	762922	762805	763071	266	FTL_0773	115	+	-	hypothetical protein, pseudogene
256	3.3	762922	762805	763071	266	FTL_0774	581	+	-	hypothetical protein, pseudogene
256	3.3	762922	762805	763071	266	FTL_0775	929	+	-	hypothetical protein, pseudogene
257	3.29	530977	530824	531153	329	FTL_0547	33	-	-	3-deoxy-D-manno-octulosonic-acid transferase
257	3.29	530977	530824	531153	329	FTL_0548	-650	-	-	putative deoxyribonucleotide triphosphate pyrophosphatase
257	3.29	530977	530824	531153	329	FTL_0549	757	+	-	pyrroline-5-carboxylate reductase
258	3.26	24982	24856	25170	314	FTL_0024	554	-	-	serine transporter
258	3.26	24982	24856	25170	314	FTL_0025	-6	-	-	hypothetical protein
258	3.26	24982	24856	25170	314	FTL_0026	498	+	-	3-hydroxyisobutyrate dehydrogenase
258	3.26	24982	24856	25170	314	FTL_0027	858	+	-	3-hydroxyisobutyrate dehydrogenase
259	3.24	892408	892261	892561	300	FTL_0917	-645	-	-	transcription-repair coupling

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

										factor
260	3.24	1257166	1256983	1257276	293	FTL_1320	-221	-	-	hypothetical protein
260	3.24	1257166	1256983	1257276	293	FTL_1321	-501	-	-	hypothetical protein
260	3.24	1257166	1256983	1257276	293	FTL_1322	-931	-	-	hypothetical protein, pseudogene
261	3.24	679132	678941	679254	313	FTL_0690	-97	-	-	Acyl-CoA synthetase (long-chain-fatty-acid--CoA ligase)
263	3.18	857285	857145	857445	300	FTL_0879	-903	-	-	beta-lactamase
264	3.18	871012	870895	871156	261	FTL_0893	-618	+	*clpX*	ATP-dependent protease ATP-binding subunit ClpX
264	3.18	871012	870895	871156	261	FTL_0894	660	+	-	DNA-binding, ATP-dependent protease La
265	3.18	689362	689283	689476	193	FTL_0701	-636	+	-	FAD binding family protein
266	3.16	124918	124809	125018	209	FTL_0126	618	-	-	hypothetical protein
266	3.16	124918	124809	125018	209	FTL_R0003	-243	+	-	16S ribosomal RNA
267	3.15	273273	273108	273407	299	FTL_0286	561	-	-	hypothetical protein
268	3.15	524821	524677	524996	319	FTL_0541	-614	+	-	hypothetical protein
268	3.15	524821	524677	524996	319	FTL_0542	64	+	-	hypothetical protein
268	3.15	524821	524677	524996	319	FTL_0543	845	+	-	hypothetical protein, pseudogene
269	3.14	1496625	1496479	1496789	310	FTL_1567	491	-	-	major facilitator transporter
269	3.14	1496625	1496479	1496789	310	FTL_1568	-380	+	-	LysR family transcriptional regulator
269	3.14	1496625	1496479	1496789	310	FTL_1569	583	+	-	phosphoglycolate phosphatase
270	3.14	415936	415806	416033	227	FTL_0445	729	-	-	hypothetical protein
270	3.14	415936	415806	416033	227	FTL_R0016	-240	+	-	16S ribosomal RNA
271	3.14	1135415	1135089	1135500	411	FTL_1185	560	-	-	macrophage growth locus subunit A
273	3.12	991590	991489	991793	304	FTL_1034	716	+	-	sulfate adenylyltransferase
274	3.12	294877	294735	295019	284	FTL_0307	868	-	*coaE*	dephospho-CoA kinase
274	3.12	294877	294735	295019	284	FTL_0308	-670	+	-	hypothetical protein
274	3.12	294877	294735	295019	284	FTL_0309	681	+	*aceE*	pyruvate dehydrogenase

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

										subunit E1
275	3.11	1558005	1557860	1558185	325	FTL_1628	-247	+	-	hypothetical protein
276	3.09	181652	181506	181833	327	FTL_0180	-46	+	-	acyltransferase
276	3.09	181652	181506	181833	327	FTL_0181	890	+	-	Type IV pili fiber building block protein
277	3.05	1042831	1042665	1042944	279	FTL_1094	-410	-	-	bifunctional methionine sulfoxide reductase B/A protein, pseudogene
277	3.05	1042831	1042665	1042944	279	FTL_1095	-717	-	-	bifunctional methionine sulfoxide reductase B/A protein, pseudogene
278	3.04	1671480	1671369	1671894	525	FTL_1740	-74	-	-	fatty acid desaturase
279	3.03	1163195	1163088	1163400	312	FTL_1212	774	-	*leuS*	leucyl-tRNA synthetase
280	3.01	1721157	1720955	1721288	333	FTL_1788	225	-	-	succinate dehydrogenase, cytochrome b556
280	3.01	1721157	1720955	1721288	333	FTL_1789	117	+	-	citrate synthase
281	3.01	1149614	1149478	1149772	294	FTL_1199	536	-	-	regulatory protein
281	3.01	1149614	1149478	1149772	294	FTL_1200	-388	+	-	cobalamin (vitamin B12) synthesis protein/P47K family protein
281	3.01	1149614	1149478	1149772	294	FTL_1201	832	+	-	ZIP metal transporter family protein
283	2.96	922748	922364	922880	516	FTL_0953	401	-	-	hypothetical protein
283	2.96	922748	922364	922880	516	FTL_0954	-457	-	-	hypothetical protein
286	2.89	772969	772847	773102	255	FTL_0785	679	-	-	GTP-binding protein
286	2.89	772969	772847	773102	255	FTL_0786	-616	+	-	N-acetylglucosamine-6-phosphate deacetylase
288	2.86	789816	789641	790009	368	FTL_0804	-897	+	-	hypothetical protein
288	2.86	789816	789641	790009	368	FTL_0805	-593	+	-	bifunctional proline dehydrogenase/pyrroline-5-carboxylate dehydrogenase
290	2.83	1727140	1727036	1727314	278	FTL_1793	-857	-	-	hypothetical protein
291	2.8	332428	332269	332559	290	FTL_0353	515	-	*isftu1*	transposase
291	2.8	332428	332269	332559	290	FTL_0354	38	-	-	hypothetical protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

291	2.8	332428	332269	332559	290	FTL_0355	-830	-	*ubiA*	4-hydroxybenzoate polyprenyltransferase
292	2.8	1438621	1438418	1438701	283	FTL_1505	415	-	*isftu1*	transposase
292	2.8	1438621	1438418	1438701	283	FTL_1506	-341	-	-	short-chain dehydrogenase
292	2.8	1438621	1438418	1438701	283	FTL_1507	-673	-	-	3-oxoacyl-ACP reductase
293	2.76	719204	719029	719326	297	FTL_0730	638	+	-	haloacid dehalogenase-like hydrolase family protein
294	2.75	4738	4555	4867	312	FTL_0004	522	-	*isftu1*	transposase
294	2.75	4738	4555	4867	312	FTL_0005	-432	+	-	Na+/H+ antiporter NHAP, fragment
296	2.73	1201295	1201131	1201416	285	FTL_1257	-937	+	-	glutathione reductase, pseudogene
296	2.73	1201295	1201131	1201416	285	FTL_1258	-27	+	-	aldo/keto reductase
296	2.73	1201295	1201131	1201416	285	FTL_1259	940	+	-	aldo/keto reductase, pseudogene
297	2.73	1490734	1490601	1490878	277	FTL_1559	-482	+	-	hypothetical protein, pseudogene
297	2.73	1490734	1490601	1490878	277	FTL_1560	232	+	-	hypothetical protein, pseudogene
297	2.73	1490734	1490601	1490878	277	FTL_1561	591	+	-	hypothetical protein, pseudogene
297	2.73	1490734	1490601	1490878	277	FTL_1562	966	+	-	hypothetical protein, pseudogene
298	2.72	1600383	1600223	1600542	319	FTL_1666	959	-	-	DNA polymerase I
298	2.72	1600383	1600223	1600542	319	FTL_1667	-105	-	*lpxK*	tetraacyldisaccharide 4'-kinase
299	2.7	1163736	1163583	1163879	296	FTL_1213	-688	-	-	hypothetical protein
301	2.7	1508322	1508222	1508504	282	FTL_1581	-409	+	-	lipoprotein
302	2.69	345992	345872	346122	250	FTL_0373	106	-	-	hypothetical protein
304	2.69	1032440	1032305	1032565	260	FTL_1079	441	-	-	helix-turn-helix family protein
304	2.69	1032440	1032305	1032565	260	FTL_1080	-109	-	-	hypothetical protein
304	2.69	1032440	1032305	1032565	260	FTL_1081	462	+	-	hypothetical protein
304	2.69	1032440	1032305	1032565	260	FTL_1082	850	+	-	hypothetical protein
305	2.68	1088738	1088610	1088849	239	FTL_1146	-549	+	-	glyceraldehyde-3-phosphate dehydrogenase

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

305	2.68	1088738	1088610	1088849	239	FTL_1147	471	+	*pgk*	phosphoglycerate kinase
306	2.67	1413433	1413292	1413552	260	FTL_1486	520	-	-	tRNA-(ms(2)io(6)a)-hydroxylase
306	2.67	1413433	1413292	1413552	260	FTL_1487	-404	+	-	uridine phosphorylase
306	2.67	1413433	1413292	1413552	260	FTL_1488	420	+	-	cytidine deaminase
306	2.67	1413433	1413292	1413552	260	FTL_R0040	-997	-	*tRNA-Lys1*	Lys tRNA
308	2.65	1725281	1725033	1725371	338	FTL_1791	609	-	-	superoxide dismutase
308	2.65	1725281	1725033	1725371	338	FTL_1792	-473	+	-	glutaredoxin-like protein
309	2.58	1311569	1311395	1311688	293	FTL_1376	768	-	-	carbohydrate kinase family protein (YjeF-related protein)
309	2.58	1311569	1311395	1311688	293	FTL_1377	-186	-	-	hypothetical protein
309	2.58	1311569	1311395	1311688	293	FTL_1378	-480	-	-	hypothetical protein, pseudogene
311	2.57	927310	927137	927417	280	FTL_0958	674	-	-	hypothetical protein
311	2.57	927310	927137	927417	280	FTL_0959	-184	-	-	Type IV pili leader peptidase and methylase.
312	2.57	1701292	1701176	1701454	278	FTL_1766	-577	+	-	hypothetical protein
312	2.57	1701292	1701176	1701454	278	FTL_1767	-134	+	-	hypothetical protein, pseudogene
312	2.57	1701292	1701176	1701454	278	FTL_1768	192	+	-	hypothetical protein, pseudogene
312	2.57	1701292	1701176	1701454	278	FTL_1769	412	+	-	hypothetical protein, pseudogene
313	2.57	1650764	1650584	1650867	283	FTL_1718	389	-	*isftu1*	transposase
313	2.57	1650764	1650584	1650867	283	FTL_1719	-398	+	-	hypothetical protein
314	2.54	579882	579744	580029	285	FTL_0594	707	+	-	UDP-glucose 4-epimerase
316	2.52	967563	967436	967700	264	FTL_1000	-871	+	-	hypothetical protein
316	2.52	967563	967436	967700	264	FTL_1001	-652	+	-	hypothetical protein
316	2.52	967563	967436	967700	264	FTL_1002	95	+	-	hypothetical protein
316	2.52	967563	967436	967700	264	FTL_1003	877	+	-	DNA polymerase III subunit epsilon
317	2.5	1397302	1397171	1397424	253	FTL_1472	-810	+	-	hypothetical protein
320	2.45	145089	144976	145247	271	FTL_0138	-838	+	-	ribonuclease II family protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

321	2.45	155170	155072	155253	181	FTL_0148	-475	+	-	Sodium/hydrogen exchanger (antiporter) family protein
324	2.37	1801898	1801771	1802110	339	FTL_1870	-696	-	-	secretion protein, pseudogene
325	2.36	1014772	1014654	1014892	238	FTL_1060	-178	-	-	D-alanyl-D-alanine carboxypeptidase
325	2.36	1014772	1014654	1014892	238	FTL_1061	335	+	-	inorganic pyrophosphatase
325	2.36	1014772	1014654	1014892	238	FTL_1062	947	+	-	3-deoxy-D-manno-octulosonate 8-phosphate phosphatase
326	2.36	193736	193570	193835	265	FTL_0192	-330	+	-	cytochrome O ubiquinol oxidase subunit I
327	2.35	1252840	1252738	1253013	275	FTL_1314	-178	-	-	hypothetical protein
327	2.35	1252840	1252738	1253013	275	FTL_1315	-658	-	-	hypothetical protein
327	2.35	1252840	1252738	1253013	275	FTL_1316	649	+	*isftu1*	transposase
329	2.33	1366085	1365906	1366213	307	FTL_1437	-982	-	-	hypothetical protein
330	2.31	1535645	1535515	1535784	269	FTL_1605	999	-	-	putative periplasmic protease
331	2.28	1077953	1077836	1078049	213	FTL_1134	-518	-	-	hypothetical protein
331	2.28	1077953	1077836	1078049	213	FTL_1135	640	+	-	hypothetical protein, pseudogene
332	2.28	758247	758084	758361	277	FTL_0767	473	-	-	hypothetical protein
333	2.27	1356870	1356796	1357042	246	FTL_1425	912	-	-	hypothetical protein
333	2.27	1356870	1356796	1357042	246	FTL_1426	234	-	-	hypothetical protein
334	2.27	1682085	1681903	1682191	288	FTL_1744	677	-	*rpoB*	DNA-directed RNA polymerase subunit beta
334	2.27	1682085	1681903	1682191	288	FTL_1745	146	-	*rplL*	50S ribosomal protein L7/L12
334	2.27	1682085	1681903	1682191	288	FTL_1746	-434	-	*rplJ*	50S ribosomal protein L10
336	2.27	482406	482248	482486	238	FTL_0496	-105	+	-	aspartate-semialdehyde dehydrogenase, pseudogene
336	2.27	482406	482248	482486	238	FTL_0497	34	+	-	aspartate-semialdehyde dehydrogenase, pseudogene
337	2.26	192689	192535	192808	273	FTL_0190	584	-	-	major facilitator transporter
337	2.26	192689	192535	192808	273	FTL_0191	-224	+	-	cytochrome O ubiquinol oxidase subunit II
339	2.24	1461920	1461757	1462035	278	FTL_1530	883	-	-	phosphopyruvate hydratase,

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

										pseudogene
339	2.24	1461920	1461757	1462035	278	FTL_1531	-476	-	*rumA*	23S rRNA 5-methyluridine methyltransferase
340	2.23	700345	700227	700512	285	FTL_0710	-956	-	-	choloylglycine hydrolase family protein
341	2.22	907509	907389	907624	235	FTL_0935	310	-	-	hypothetical protein
341	2.22	907509	907389	907624	235	FTL_0936	-67	-	-	hypothetical protein
341	2.22	907509	907389	907624	235	FTL_0937	346	+	-	hypothetical protein
343	2.21	1564604	1564474	1564719	245	FTL_1634	264	-	-	LysR family transcriptional regulator
343	2.21	1564604	1564474	1564719	245	FTL_1635	274	+	-	chitinase, fragment
344	2.21	1285186	1285070	1285332	262	FTL_1350	-171	-	-	glycyl-tRNA synthetase subunit beta
344	2.21	1285186	1285070	1285332	262	FTL_1351	364	+	-	TatD related DNAse family protein
345	2.19	1528138	1527990	1528260	270	FTL_1598	-951	+	*argS*	arginyl-tRNA synthetase
346	2.19	553143	553056	553273	217	FTL_0574	-531	+	-	hypothetical protein
347	2.17	759551	759395	759632	237	FTL_0768	-212	-	-	GTP binding translational elongation factor Tu and G family protein
347	2.17	759551	759395	759632	237	FTL_0769	938	+	-	hypothetical protein
348	2.17	1571866	1571746	1571994	248	FTL_1640	-549	+	-	amino acid transporter protein, fragment
348	2.17	1571866	1571746	1571994	248	FTL_1641	966	+	-	hypothetical protein
349	2.17	1320100	1319942	1320171	229	FTL_1389	223	-	-	nicotinate-nucleotide pyrophosphorylase
349	2.17	1320100	1319942	1320171	229	FTL_1390	-811	-	-	quinolinate synthetase
350	2.15	1658266	1658167	1658435	268	FTL_1726	-770	+	-	DNA topoisomerase IV subunit B
351	2.15	1492994	1492888	1493140	252	FTL_1563	-639	+	-	hypothetical protein, pseudogene
351	2.15	1492994	1492888	1493140	252	FTL_1564	101	-	-	hypothetical protein, pseudogene
351	2.15	1492994	1492888	1493140	252	FTL_1565	-395	-	-	hypothetical protein, pseudogene

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

354	2.14	1596974	1596858	1597083	225	FTL_1665	263	-	-	pantothenate kinase
355	2.11	729652	729576	729734	158	FTL_0739	-106	-	-	tRNA uridine 5-carboxymethylaminomethyl modification protein GidA
355	2.11	729652	729576	729734	158	FTL_0740	249	+	-	hypothetical protein
356	2.11	12854	12800	12910	110	FTL_0016	372	+	-	phosphate acetyltransferase
357	2.1	994962	994801	995082	281	FTL_1037	-199	+	-	hypothetical protein
357	2.1	994962	994801	995082	281	FTL_1038	419	+	-	hypothetical protein
359	2.08	1249676	1249597	1249802	205	FTL_1311	-334	-	*pyrG*	CTP synthetase
359	2.08	1249676	1249597	1249802	205	FTL_1312	647	+	-	S-transferase
360	2.08	1740847	1740723	1740930	207	FTL_1806	968	-	-	major facilitator transporter
360	2.08	1740847	1740723	1740930	207	FTL_1807	-304	-	*hisS*	histidyl-tRNA synthetase
360	2.08	1740847	1740723	1740930	207	FTL_1808	-743	-	-	ribosome-binding factor A
361	2.07	1330059	1329751	1330269	518	FTL_1399	112	-	-	3-deoxy-manno-octulosonate cytidylyltransferase
361	2.07	1330059	1329751	1330269	518	FTL_1400	-129	-	-	hypothetical protein
361	2.07	1330059	1329751	1330269	518	FTL_1401	-726	-	-	hypothetical protein
362	2.06	1082740	1082618	1082846	228	FTL_1139	643	-	-	3-oxoacyl-(acyl-carrier-protein) reductase
363	2.05	23320	23188	23491	303	FTL_R0001	174	-	*ssrA*	-
364	2.05	1347578	1347490	1347752	262	FTL_1417	230	-	-	major facilitator transporter
364	2.05	1347578	1347490	1347752	262	FTL_R0035	-20	-	*tRNA-Leu4*	Leu tRNA
364	2.05	1347578	1347490	1347752	262	FTL_R0036	-102	-	*tRNA-His1*	His tRNA
364	2.05	1347578	1347490	1347752	262	FTL_R0037	-206	-	*tRNA-Arg2*	Arg tRNA
364	2.05	1347578	1347490	1347752	262	FTL_R0038	-291	-	*tRNA-Pro1*	Pro tRNA
364	2.05	1347578	1347490	1347752	262	FTL_R0039	-384	-	*tRNA-Phe1*	Phe tRNA
366	2.04	904393	904314	904604	290	FTL_0932	-231	+	*ruvA*	Holliday junction DNA helicase RuvA
366	2.04	904393	904314	904604	290	FTL_0933	445	+	-	DNA recombination protein RmuC family protein
367	2.03	451221	451101	451329	228	FTL_0475	857	+	-	lipoprotein releasing system, subunit D, ABC transporter

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

										ATP-binding protein
368	2.03	702271	702143	702392	249	FTL_0711	-474	+	-	proton-dependent oligopeptide transport (POT) family protein
368	2.03	702271	702143	702392	249	FTL_0712	210	+	-	hypothetical protein
369	2.02	589201	589023	589328	305	FTL_0601	-219	+	-	sugar transamine/perosamine synthetase
369	2.02	589201	589023	589328	305	FTL_0602	860	+	-	hypothetical protein
371	2.02	1709330	1709173	1709439	266	FTL_1776	238	-	-	hypothetical protein
371	2.02	1709330	1709173	1709439	266	FTL_1777	-231	-	-	hypothetical protein, pseudogene
371	2.02	1709330	1709173	1709439	266	FTL_1778	-685	-	-	hypothetical protein, pseudogene
372	2.01	1407638	1407481	1407732	251	FTL_1481	-204	+	-	YjgP/YjgQ family permease
373	2	1518451	1518392	1518657	265	FTL_1589	734	-	*isftu2*	transposase
373	2	1518451	1518392	1518657	265	FTL_1590	-533	-	-	hypothetical protein
375	1.98	549901	549807	550034	227	FTL_0570	-960	+	-	hypothetical protein
375	1.98	549901	549807	550034	227	FTL_0571	-384	+	-	hypothetical protein
375	1.98	549901	549807	550034	227	FTL_0572	281	+	-	hypothetical protein
376	1.98	606642	606541	606826	285	FTL_0616	-568	+	-	DNA-directed RNA polymerase subunit alpha
376	1.98	606642	606541	606826	285	FTL_R0022	394	+	*tRNA-Leu1*	Leu tRNA
377	1.98	1140196	1140168	1140278	110	FTL_1192	876	+	-	chaperone protein DnaJ
379	1.95	1239985	1239907	1240049	142	FTL_1301	-898	+	*isftu2*	transposase
379	1.95	1239985	1239907	1240049	142	FTL_1302	-184	+	-	phenol hydroxylase
379	1.95	1239985	1239907	1240049	142	FTL_1303	669	+	*rpmE*	50S ribosomal protein L31
381	1.94	519355	519299	519438	139	FTL_0536	828	+	-	outer membrane protein OmpH
382	1.94	478768	478676	478964	288	FTL_0493	309	+	-	hypothetical protein
384	1.92	1510652	1510518	1510770	252	FTL_1582	490	-	-	putrescine-binding periplasmic protein
384	1.92	1510652	1510518	1510770	252	FTL_1583	-406	+	-	glutamate:gamma-aminobutyric acid antiporter family protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

385	1.92	47184	47001	47273	272	FTL_0044	-356	-	-	transglutaminase
385	1.92	47184	47001	47273	272	FTL_0045	776	+	-	orotidine 5'-phosphate decarboxylase
386	1.91	365490	365389	365588	199	FTL_0395	-866	+	-	phosphoribosylaminoimidazole synthetase
386	1.91	365490	365389	365588	199	FTL_0396	177	+	-	fusion protein PurC/PurD
389	1.88	1634528	1634423	1634626	203	FTL_1703	-592	-	-	amino acid transporter
390	1.88	566688	566636	566733	97	FTL_0585	-474	-	*fadE*	acyl-CoA dehydrogenase
392	1.86	1753364	1753205	1753490	285	FTL_1818	154	-	-	NADH dehydrogenase I subunit M
393	1.85	1273540	1273432	1273643	211	FTL_1337	259	-	-	prephenate dehydratase, pseudogene
393	1.85	1273540	1273432	1273643	211	FTL_1338	-956	-	-	alanine racemase
394	1.85	1210439	1210267	1210545	278	FTL_1267	-614	+	-	hypothetical protein
394	1.85	1210439	1210267	1210545	278	FTL_1268	34	+	-	hypothetical protein, pseudogene
394	1.85	1210439	1210267	1210545	278	FTL_1269	137	+	-	hypothetical protein, pseudogene
394	1.85	1210439	1210267	1210545	278	FTL_1270	456	+	-	hypothetical protein, pseudogene
395	1.84	1850386	1850212	1850467	255	FTL_1916	-602	-	-	competence-like protein
395	1.84	1850386	1850212	1850467	255	FTL_1917	766	+	-	30S ribosomal protein S6
396	1.84	436178	436076	436236	160	FTL_0462	-406	+	-	DNA topoisomerase IV subunit A
397	1.84	1151667	1151562	1151752	190	FTL_1202	-536	-	-	hypothetical protein
398	1.83	583280	583161	583394	233	FTL_0596	-801	+	-	UDP-glucose/GDP-mannose dehydrogenase
399	1.83	1636280	1636173	1636414	241	FTL_1704	245	-	-	hypothetical protein
399	1.83	1636280	1636173	1636414	241	FTL_1705	-123	+	-	cell division protein
402	1.82	227280	227206	227330	124	FTL_0226	-466	+	*pyrH*	uridylate kinase
402	1.82	227280	227206	227330	124	FTL_0227	313	+	*frr*	ribosome recycling factor
402	1.82	227280	227206	227330	124	FTL_0228	954	+	-	undecaprenyl pyrophosphate synthase
403	1.81	242049	241856	242155	299	FTL_0251	-892	+	*rplF*	50S ribosomal protein L6

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

403	1.81	242049	241856	242155	299	FTL_0252	-333	+	*rplR*	50S ribosomal protein L18
403	1.81	242049	241856	242155	299	FTL_0253	48	+	*rpsE*	30S ribosomal protein S5
403	1.81	242049	241856	242155	299	FTL_0254	555	+	*rpmD*	50S ribosomal protein L30
403	1.81	242049	241856	242155	299	FTL_0255	747	+	*rplO*	50S ribosomal protein L15
404	1.81	1766132	1765987	1766224	237	FTL_1832	-608	+	-	hypothetical protein
405	1.81	1291025	1290938	1291151	213	FTL_1356	871	-	-	TatD related DNAse family protein, pseudogene
405	1.81	1291025	1290938	1291151	213	FTL_1357	-114	-	-	UTP--glucose-1-phosphate uridylyltransferase
406	1.79	1071842	1071725	1071952	227	FTL_1125	596	-	-	transcriptional regulator
406	1.79	1071842	1071725	1071952	227	FTL_1126	366	-	-	transcriptional regulator
407	1.78	162164	162037	162304	267	FTL_0157	-263	-	-	hypothetical protein
407	1.78	162164	162037	162304	267	FTL_0158	570	+	-	acid phosphatase
408	1.78	102675	102522	102818	296	FTL_0108	611	-	-	hypothetical protein
408	1.78	102675	102522	102818	296	FTL_0109	-518	+	-	hypothetical protein
408	1.78	102675	102522	102818	296	FTL_0110	-107	+	-	hypothetical protein
408	1.78	102675	102522	102818	296	FTL_0111	177	+	-	intracellular growth locus, subunit A
408	1.78	102675	102522	102818	296	FTL_0112	745	+	-	intracellular growth locus, subunit B
409	1.77	1735589	1735499	1735721	222	FTL_1801	867	-	-	F0F1 ATP synthase subunit A
409	1.77	1735589	1735499	1735721	222	FTL_1802	383	-	-	hypothetical protein
409	1.77	1735589	1735499	1735721	222	FTL_1803	-306	+	-	major facilitator transporter
410	1.77	196759	196685	196918	233	FTL_0194	-722	+	-	cytochrome O ubiquinol oxidase subunit IV
410	1.77	196759	196685	196918	233	FTL_0195	-359	+	-	protoheme IX farnesyltransferase
412	1.76	219768	219657	219891	234	FTL_0219	165	+	-	hypothetical protein
414	1.75	874011	873890	874126	236	FTL_0895	72	+	-	histone-like protein HU form B
414	1.75	874011	873890	874126	236	FTL_0896	457	+	-	hypothetical protein
415	1.75	278357	278234	278506	272	FTL_0292	429	-	-	hypothetical protein, pseudogene

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

415	1.75	278357	278234	278506	272	FTL_0293	-426	-	-	preprotein translocase subunit SecB
417	1.75	641215	641155	641273	118	FTL_0659	-559	-	-	cell division protein
417	1.75	641215	641155	641273	118	FTL_0660	668	+	-	ferrous iron transport protein A
417	1.75	641215	641155	641273	118	FTL_0661	909	+	-	hypothetical protein
418	1.74	1370950	1370790	1371000	210	FTL_1442	797	-	-	enoyl-[acyl-carrier-protein] reductase (NADH)
418	1.74	1370950	1370790	1371000	210	FTL_1443	-511	+	-	hypothetical protein
418	1.74	1370950	1370790	1371000	210	FTL_1444	216	+	-	hypothetical protein
418	1.74	1370950	1370790	1371000	210	FTL_1445	945	+	*isftu2*	transposase
420	1.73	318077	317958	318181	223	FTL_0335	142	+	-	lipoprotein
420	1.73	318077	317958	318181	223	FTL_0336	406	+	-	peptidoglycan-associated lipoprotein
421	1.73	335347	335221	335443	222	FTL_0358	397	-	-	hypothetical protein
421	1.73	335347	335221	335443	222	FTL_0359	-610	-	-	Type IV pili fiber building block protein
422	1.72	412564	412462	412671	209	FTL_0443	423	-	-	major facilitator transporter
423	1.72	492614	492449	492732	283	FTL_0507	783	-	*pyrE*	orotate phosphoribosyltransferase
423	1.72	492614	492449	492732	283	FTL_0508	-623	+	-	UDP-N-acetylmuramate--L-alanyl-gamma-D-glutamyl-meso-diaminopimelate ligase
423	1.72	492614	492449	492732	283	FTL_0509	744	+	-	tRNA/rRNA methyltransferase
424	1.71	281934	281846	282040	194	FTL_0296	594	+	-	hypothetical protein
425	1.71	235202	235157	235252	95	FTL_0236	-790	+	*rplC*	50S ribosomal protein L3
425	1.71	235202	235157	235252	95	FTL_0237	-125	+	*rplD*	50S ribosomal protein L4
429	1.7	224588	224537	224688	151	FTL_0222	205	-	-	hypothetical protein
429	1.7	224588	224537	224688	151	FTL_0223	-419	-	-	dihydrofolate reductase type I
429	1.7	224588	224537	224688	151	FTL_0224	612	+	*rpsB*	30S ribosomal protein S2
431	1.68	375253	375178	375368	190	FTL_0406	-841	+	-	hypothetical protein
431	1.68	375253	375178	375368	190	FTL_0407	-261	+	-	2-polyprenylphenol 6-hydroxylase
433	1.67	444569	444451	444714	263	FTL_0469	504	-	-	hypothetical protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

433	1.67	444569	444451	444714	263	FTL_0470	-151	+	-	hypothetical protein
433	1.67	444569	444451	444714	263	FTL_0471	589	+	-	hypothetical protein
434	1.67	1554027	1553987	1554064	77	FTL_1622	837	-	-	multidrug transporter (tetracycline resistance protein)
434	1.67	1554027	1553987	1554064	77	FTL_1623	-289	-	-	hypothetical protein
434	1.67	1554027	1553987	1554064	77	FTL_1624	561	+	-	major facilitator transporter
435	1.67	549229	549068	549334	266	FTL_0569	-942	+	-	hypothetical protein
436	1.67	1532892	1532783	1533026	243	FTL_1602	478	-	-	delta-aminolevulinic acid dehydratase
436	1.67	1532892	1532783	1533026	243	FTL_1603	192	-	-	RNA-binding protein
436	1.67	1532892	1532783	1533026	243	FTL_1604	-730	-	-	DNA polymerase III, delta prime subunit
437	1.67	481872	481826	482003	177	FTL_0494	879	-	-	aspartate-semialdehyde dehydrogenase
437	1.67	481872	481826	482003	177	FTL_0495	-589	+	-	aspartate-semialdehyde dehydrogenase, pseudogene
438	1.66	515630	515566	515779	213	FTL_0534	876	+	-	1-deoxy-D-xylulose 5-phosphate reductoisomerase
441	1.66	313399	313316	313508	192	FTL_0328	-143	+	-	chorismate mutase
441	1.66	313399	313316	313508	192	FTL_0329	234	+	-	CDP-alcohol phosphatidyltransferase
442	1.65	900158	900100	900193	93	FTL_0926	533	-	-	Ferritin-like protein
442	1.65	900158	900100	900193	93	FTL_0927	-327	+	-	lipoyl synthase
442	1.65	900158	900100	900193	93	FTL_0928	686	+	-	isoprenoid biosynthesis protein with amidotransferase-like domain
443	1.65	1607754	1607618	1607824	206	FTL_1673	918	+	-	major facilitator transporter
444	1.65	29987	29935	30048	113	FTL_0029	-731	-	*carB*	carbamoyl phosphate synthase large subunit
445	1.65	932020	931936	932085	149	FTL_0963	334	-	-	proton-dependent oligopeptide transport (POT) family protein
446	1.64	1026825	1026743	1026899	156	FTL_1073	149	+	-	hypothetical protein
446	1.64	1026825	1026743	1026899	156	FTL_1074	506	+	-	GDSL-like lipase/acylhydrolase family

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

										protein
447	1.64	513280	513210	513351	141	FTL_0530	687	-	-	hypothetical protein
447	1.64	513280	513210	513351	141	FTL_0531	225	-	-	hypothetical protein
448	1.64	1097318	1097170	1097464	294	FTL_1154	615	-	-	phosphoglycerate kinase, pseudogene
448	1.64	1097318	1097170	1097464	294	FTL_1155	-522	+	-	hypothetical protein
448	1.64	1097318	1097170	1097464	294	FTL_1156	-111	+	-	hypothetical protein
448	1.64	1097318	1097170	1097464	294	FTL_1157	173	+	-	intracellular growth locus, subunit A
448	1.64	1097318	1097170	1097464	294	FTL_1158	741	+	-	intracellular growth locus, subunit B
451	1.63	832893	832811	832995	184	FTL_0850	-888	+	-	hypothetical protein
451	1.63	832893	832811	832995	184	FTL_0851	-334	+	-	RNA polymerase factor sigma-32
451	1.63	832893	832811	832995	184	FTL_0852	635	+	-	3-phosphoshikimate 1-carboxyvinyltransferase
452	1.62	1504494	1504361	1504568	207	FTL_1576	-701	-	-	DNA mismatch repair protein
453	1.62	214321	214223	214434	211	FTL_0213	-394	+	-	hypothetical protein
454	1.62	555726	555599	555862	263	FTL_0575	866	-	*isftu2*	transposase
454	1.62	555726	555599	555862	263	FTL_0576	-572	+	-	hypothetical protein
455	1.61	742176	742109	742314	205	FTL_0750	-657	+	-	major facilitator transporter, pseudogene
455	1.61	742176	742109	742314	205	FTL_0751	566	+	-	lipoprotein
456	1.61	1852038	1851850	1852110	260	FTL_1918	-92	+	-	30S ribosomal protein S6 modification protein-like protein
456	1.61	1852038	1851850	1852110	260	FTL_1919	927	+	-	30S ribosomal protein S6 modification protein-like protein, pseudogene
458	1.6	189310	189229	189418	189	FTL_0188	790	-	-	cytochrome d terminal oxidase, polypeptide subunit II
458	1.6	189310	189229	189418	189	FTL_0189	-975	-	-	cytochrome d terminal oxidase, polypeptide subunit I
460	1.6	1075373	1075301	1075446	145	FTL_1131	127	-	-	Sun protein

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

461	1.59	1732505	1732331	1732595	264	FTL_1797	9	-	-	F0F1 ATP synthase subunit alpha
461	1.59	1732505	1732331	1732595	264	FTL_1798	-534	-	-	F0F1 ATP synthase subunit delta
462	1.59	325149	325043	325269	226	FTL_0344	-69	+	-	hypothetical protein
462	1.59	325149	325043	325269	226	FTL_0345	194	+	-	bile acid symporter family protein
463	1.59	739073	738973	739154	181	FTL_0748	-138	+	-	major facilitator transporter
464	1.58	545697	545583	545788	205	FTL_0564	-577	-	-	hypothetical protein, pseudogene
464	1.58	545697	545583	545788	205	FTL_0565	-945	-	-	hypothetical protein, pseudogene
466	1.57	748861	748825	748905	80	FTL_0757	180	+	-	hypothetical protein, pseudogene
466	1.57	748861	748825	748905	80	FTL_0758	585	+	-	hypothetical protein, pseudogene
467	1.57	716128	716053	716195	142	FTL_0726	-882	+	-	2-octaprenyl-6-methoxyphenyl hydroxylase
467	1.57	716128	716053	716195	142	FTL_0727	319	+	-	monooxygenase family protein
468	1.56	839037	838935	839133	198	FTL_0857	518	-	-	hypothetical protein
469	1.55	1863237	1863162	1863436	274	FTL_1931	-168	+	-	hypoxanthine-guanine phosphoribosyltransferase
469	1.55	1863237	1863162	1863436	274	FTL_1932	-557	-	-	hypoxanthine-guanine phosphoribosyltransferase, pseudogene
470	1.54	1885699	1885551	1885812	261	FTL_1956	-140	-	*pepN*	aminopeptidase N
470	1.54	1885699	1885551	1885812	261	FTL_1957	342	+	-	heat shock protein
471	1.54	1699043	1698994	1699132	138	FTL_1763	917	-	-	hypothetical protein
471	1.54	1699043	1698994	1699132	138	FTL_1764	-399	+	-	hypothetical protein, pseudogene
471	1.54	1699043	1698994	1699132	138	FTL_1765	699	+	-	cytochrome oxidase bd-II subunit II
472	1.54	706959	706873	707075	202	FTL_0715	693	-	*thyA*	thymidylate synthase
472	1.54	706959	706873	707075	202	FTL_0716	-98	-	-	prolipoprotein diacylglyceryl transferase

Table S19. Genes associated* with IclR (FTL_1364) ChIP-Seq peaks, continued

474	1.53	1694968	1694887	1695118	231	FTL_1759	180	-	-	hypothetical protein
474	1.53	1694968	1694887	1695118	231	FTL_1760	-351	-	-	hypothetical protein
474	1.53	1694968	1694887	1695118	231	FTL_1761	-714	-	-	hypothetical protein
474	1.53	1694968	1694887	1695118	231	FTL_1762	853	+	-	sensor histidine kinase
475	1.53	169122	169023	169222	199	FTL_0164	820	-	*isftu1*	transposase
475	1.53	169122	169023	169222	199	FTL_0165	576	-	-	dicarboxylate MFS transporter
475	1.53	169122	169023	169222	199	FTL_0166	-376	-	-	universal stress protein

*Gene are referred to as associated if the annotated translational start site is within I kb upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[1]If peak number is not reported, no genes were identified with a translational start site within I kb upstream to 1 kb downstream of the site of ChIP-Seq peak maximum.

[2]Site of maximum ChIP-Seq peak enrichment

[3]Start of ChIP-Seq peak

[4]End of ChIP-Seq peak

[5]Distance is measured as bp from the ChIP-Seq peak maximum to the annotated translational start site.